Studying Some of Immunological Parameters of doges that Toxocara Infections in Saladinprovince

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Abstract

The study examined 60 stray dogs from both sexes and aged 3 months - 3 years old, included 30 stray dogs and 30 dogs clinically normal, served as a control group. Toxocara infection has detected in the College of veterinary medicine Health Center in Tikrit city. Clinical signs were observed, the study groups divided to two groups one of these groups infected by the parasites of the study and other group was control the result appear immunological changes for protein levels in study groups, represented with significant differences was reported between toxocariasis group and healthy control in terms of Complement 3 proteins (33.21 ±14.18726, 35.57±15.56254), Complement 4 proteins (268.5±14.34, 146.06±17.83) although increased complement 3 level in toxocariasis group compared with healthy control, While the Complement 4 reported no significant differences increased between toxocariasis group and healthy control group. The immunoglobulins was reported highly significant differences in IgM (297.4±14.18726)in toxocariasis group compared with healthy control group (159 ±15.56), While revealed no significant decrease levels of IgG of toxocariasis group (760 ±14.34) compared with healthy control group (843±17.83). *ToxocaraCanis* induces an early immunological response that detected by presence of these antibodies and immunological markers against T.canis infection.

Keywords: *Complement 3, Complement 4, Immunoglobulin G, Immunoglobulin M, Toxocara Cainis.*

Introduction

Dogs and cats are the most important animal hosts for toxocariasis, especially in developing countries where most cats and dogs have access to public parks and playgrounds, serving as the main source of soil contamination, and posing a huge risk of human exposure to infective eggs 17. Toxocariasis is a highly prevalent parasitic disease in the tropical regions of the world, with its impact on public health being typically underestimated[1–2]. Toxocariasis is a zoonotic disease usually caused by dog and cat roundworms, Toxocaracanis and T. cati. Detection and diagnosis is difficult in paratenic and accidental hosts. The roundworm Toxocara is a perfect example of a parasite moving from wild canids to their domestic counterparts and to humans[3,4]. Dogs or cats, especially in low-income and rural regions, play important roles in the transmission of Toxocara spp. through environmental contamination, which spreads the infection to humans[3]. Toxocara parasite has a notorious tendency to cause extraintestinal pathologies[5,6]. Indeed, toxocariasis includes four clinical forms, which can lead to serious health consequences[5,7,8]. The newly sequenced genome of T. canis along with transcriptomic analysis has allowed an in-depth characterization of this organism's molecular characteristics[9]. Also, knowledge of the parasite's genetic diversity has been improved and new diagnostic markers have been discovered [10, 9–11]. Indeed, evidence from recent investigations suggests that human toxocariasis is seriously neglected because limited attention has been paid to its prevention, treatment, and surveillance and because it is a non-notifiable disease[12–13]. The clinical symptoms of toxocariasis may vary from asymptomatic infection to localized symptoms (ocular and neurological) or severe systemic infection (visceral larva migrans), which is commonly complicated by blood eosinophilia(17) preposition many infections are asymptomatic and thus can be misdiagnosed[14]. Phylogenetic analyses based on the sequences of the nuclear ribosomal DNA(rDNA),

showed that Toxocara spp. form a distinct clade, in relation to their definitive hosts, which is separate from Ascaris spp.[15]. This syndrome relates to the migration of *T canis* larvae in CNS and subsequent induction of meningitis, encephalitis, cerebral vasculitis, or myelitis, usually associated with relatively non-specific clinical symptoms (eg, fever and headache)(16).

The aim of this study is to investigate the prevalence of toxocara parasite in study doges insalahdinprevence province with its immunological effects.

Materials And Method

Specimen Collection and Preparation: Serum preparation: Peripheral blood samples from cases and controls were collected by venipuncture using vacuum tubes (Vacutainer). Whole blood and plasma aliquots were obtained and stored at 4°C and -20°C, respectively until analyzed, and we use deep frozen or fresh serum samples. The specimens was obtained by Lode, The specimen after centrifugation was overstocked for up to 48 hours at 2-8°C before assay and for along storage peroid,. The samples thawing and repeated freezing must be impossible.

Detectionof C3 and C4 concentration: The radioimmuno diffusion microplates were embosomed for 24 to 72 h at room temperature. These technique essentially of[22]. Calibrating viewer was used to gauged the distance of the ring,this technique is called Mancini method. In this reaction Ag-Abspread on in semisolid phase,where antigen aspur will form into these phase. The reactants spread toward each other on the semi solid phase even they concurs, The distance of the ring is a calculated of antigen amount, the ring diameter microplates are read after of incubation 24-72 hrs[13].

Detection of IgG and IgM concentration: The examined protein, diffusing in agarose gel containing aspecific antibody will form an immune-complex, visible as aring around the well. The ring diameter is direct proportional to concentration of the analysed protein. The proportion corresponds to the diffusion time. in fact, at the end (72h), the square of diameter will be in linear proportion to the concentration of the sample.fill the wells with 5µl of sample and wait it has been completely adsorbing before handling the plate. close the plate and place it in amoist chamber for (72h)[13].

Statistical Analysis: The data analyzed by SPSS program(statistical package for social science) version**v**[15]. Quantitative variables were represented as mean ±SD. P-value less than 0.05 (< 0.05) consider statistically significant. The relationship between studied variables was assessed by using Spearman correlation.

Results

The present study was designed to estimate some immunological biomarkers (C3, C4) in studied groups; 30 Toxocariasis patients and 10 healthy controls. The association between circulating levels of these biomarkers were investigated.

The complement protein calculated by using method encompass radially diffusing of antigen that found in well through agarose gel including monoclonal Ab. Intricate of Ag-Ab are configured under the circumstances proportioned to these reactions will constitutes ring that configured made of reactions. The Diameter of ring will proportion suited ejective between the diameter and concentration in accordance with data sheet that provided with the kit. The Diameter of ring will proportion. (Table, 2) display demographic characteristics of the patients with toxocara infection, as well as relationship between disease and different variables.

Table (1): Demographic profile of 30 patients with Toxocariasis data include information about the, age, gender, disease duration residence, IgG, IgM.

Patient characteristics	Value	Probability
Age (years):	(3 mon.-3) years	-
Sex:	(Male/Female	-
IgG (%)		
N: No (%)	20 (84.2)	P<0.05*
P: No (%)	10 (15.8)	

Patient characteristics	Value	Probability
IgM(%):		
N: No (%)	10 (39.5)	P<0.05*
P: No (%)	20(60.5)	

IgG: Immunoglobulin gama ; IgM: Immunoglobulin meuosignificant: p<0.05(Chi square);* Significant: p<0.05(Chi square)

Table (2): C3, C4 Concentration in studied group

Characters	Patients Mean ±SE No.25	Healthy control Mean ±SE No.15	P value
C3/Mean ±SE	268.5±14.34	146.06±17.83	P≥0.05
C4/Mean ±SE	33.21 ±14.18726	35.57±15.56254	P≥0.05

HC: Healthy control; C: complement;; p<0.05(t test); Highly significant; P<0.05: Significant; P≥ 0.05; No-significant.

Characteristics of patients with Toxocarasis and healthy control are detailed in (Table,2). Showed Significant decreace in C4 concentration compared with healthy control which non significant level (P>0.05),While C3 protein showed Significant increace in concentration compared with healthy control in significant level (P>0.05).

Table (3): IgG, IgM Concentration in studied group

Characters	Patients Mean ±SE No.25	Healthy control Mean ±SE No.15	P value
IgG/Mean ±SE	760 ±14.34	843±17.83	P≥0.05
IgM/Mean ±SE	297.4±14.18726	159 ±15.56	P≥0.05

HC: Healthy control; IgG: immunoglobulin gama; IgM: immunoglobulin moderate p<0.05(t test); Highly significant; P<0.05: Significant; P≥ 0.05; No-significant.

Characteristics of patients with Toxocarasis and healthy control are detailed in (Table 3), Showed Significant increase in IgG concentration compared with healthy control which non significant level (P>0.05). While IgM showed Significant increace in concentration compared with healthy control in significant level (P>0.05).

Discussion

The present Study was desiged to correlation between some of immunological biomarkers (C3, C4, IgG, IgM). In studied groups infected stray cats showed a number of clinical signs include diarrhea, loss of appetite and wasting, and due to the diagnosis of infection with *Toxocaracanis* parasite. The infection rate appeared in this parasite high in stray dogs, which may be the reason for stray dogs spread in different regions and lack of attention with unhealthy culture conditions, which increases the chances of exposure to parasitic infection and other diseases. The presence of anti-T. canis antibodies from T. canis-infected sera were showed Significant decreased in IgG concentration compared with healthy control which non-significant level this level exhibited detectable amounts of IgG in sera at 14 days post-infection, While IgM showed Significant increace in concentration compared with healthy control in in significant level(P>0.05), The levels of circulating antigen were highest during the first week of infection, The production of specific anti-*T. canis* antibodies was increased with significance the lowest detectable levels were observed at 3 months post-infection T. canis infection exhibited detectable amounts of IgG in sera at 14 days post-infection this results aggregated with the same study[1]**.** No significant differences was reported between Toxocariasis group and healthy control in terms of Complement 4 proteins, Complement 3 proteins although increased complement level in Toxocarisis group compared with healthy control in in significant level(P>0.05), differences were significant and highly significant in terms of C3, and run up serum echelons of complement 3 are combination with acute phase of inflammatory interactions, Whereas C3 and C4 are completely inactivated by removal of the C-terminal arginine, C5a retains approximately 10% of its chemotactic activity, which may explain the partial inactivation of chemotaxisinthis infection

[5]. Complements and Some serological markers levels were elevated in Toxocariasis group with Healthy controle, But some of these decline Compared with healthy control however, The above results revealed that in the variations observed in the time of detection and the concentration of antibodies in sera may have been due to factors such as the species of the experimental host, strain used, age of the experimental host, sex of the animal[14].

Ethical Clearance: The Research Ethical Committee at scientific research by ethical approval of both environmental and health and higher education and scientific research ministries in Iraq.

Conflict of Interest: The authors declare that they have no conflict of interest.

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