RESEARCH ARTICLE
Journal of Biometry Studies (2023) 3(1): 12-16
DOI: 10.29329/JofBS.2023.501.03
www.biometrystudies.com

Journal of Biometry Studies

PRENSIP
www.prensip.gen.tr

Biochemical indicators of blood serum and features of toxoplasmosis clinical manifestations in dogs

Volodymyr KUSTUROV[1,*], Mykhailo BROSHKOV[1], Vasyl NAIDA[1]

[1]Odesa State Agrarian University, Faculty of Veterinary Medicine, Department of Physiology, Pathophysiology and Biochemistry, Odesa/UKRAINE

[*]Corresponding author: bu.dasha.bu@gmail.com

Received: 26/02/2023, Accepted: 19/06/2023

Abstract

Traditional approaches to the diagnosis of toxoplasmosis include etiological, immunological and imaging methods. However, veterinarians lack clinical markers that would be the basis for testing for toxoplasmosis. Authors found that for toxoplasmosis, the serum of dogs has a high content of ALT enzymes in 68.5% and AST in 91% of cases. In 39.7%, toxoplasmosis is associated with skin lesions and in 20.5% with lesions of the nervous system. In this article, authors summarize that in circumstances of existance of skin and nervous system lesions and a high content of AlAT and AsAT enzymes (according to the results of biochemical tests of blood serum) are found in dogs in veterinary clinics, - a test for toxoplasmosis is recommended. The obtained data create a basis for further research in understanding the pathogenetic mechanisms of toxoplasmosis clinical signs in dogs development. These studies will contribute to a better understanding of the epidemiology, prevention and control of toxoplasmosis.

Keywords: Toxoplasmosis, Clinical manifestation of toxoplasmosis, Biochemical profile of blood serum

Please cite this article as follows:

Kusturov, V., Broshkov, M., & Naida, V. (2023). Biochemical Indicators of Blood Serum and Features of Toxoplasmosis Clinical Manifestations in Dogs. *Journal of Biometry Studies*, *3*(1), 12-16. https://doi.org/10.29329/JofBS.2023.501.03

1. Introduction

Toxoplasmosis, caused by the obligate intracellular protozoan *Toxoplasma gondii*, is an important zoonosis of medical and veterinary importance worldwide. Although many studies of *Toxoplasma gondii* and toxoplasmosis infections in dogs are available in the literature, there are still gaps in features of diagnosis. All non-feline animals, including dogs and humans, are intermediate hosts in biological cycle of toxoplasmosis. There are three stages in the life cycle of protozoa, which explains their biological success. First, tachyzoites multiply actively in tissues, quickly spread to almost all organs and cause most of the pathology. When they reach certain tissues (central nervous system, muscles, and internal organs), they transform into bradyzoites, which remain latent in cyst form. It leads to chronic lifelong infection until the definitive host ingests the tissue. (Tenter et al., 2000) Clinical toxoplasmosis in dogs has a wide spectrum of manifestations, ranging from general symptoms such as fever and dyspnea to more specific signs including nervous, respiratory, skin and ocular signs (Calero-Bernal & Gennari, 2019). Timely detection of toxoplasmosis in dogs in the conditions of veterinary clinics is usually difficult. First, it is associated with various clinical manifestations in dogs. As a rule, the manifestation of an acute form has no specificity, therefore the therapeutic approach is directed at the treatment of one or another organ which function has been disrupted. (Liu et al., 2015) Secondly, the insufficient awareness of veterinarians regarding the spread of this disease and the peculiarities of its course in the body of animals does not motivate them to consider toxoplasmosis as an etiological factor. Thirdly, even in the case of establishing a diagnosis of toxoplasmosis, there is no sufficient experimental basis for therapeutic approaches and individual selection of pharmacological agents for each individual case. (Robert-Gangneux & Dardé, 2012). It should also be noted that due

to untimely diagnosis of toxoplasmosis in domestic animals, a chronic form of the disease develops. It is associated with the intracellular placement of the pathogen (endozooid) and, accordingly, the impossibility of its complete elimination from the organism. (Al-Malki, 2021).

Biochemical indicators are the most important physiological tools that reveal basic information regarding the diagnosis and prognosis of any disease (Lashari et al., 2018). The liver is most severely infected. As a result of the reproduction of toxoplasmas, scientists observed a number of pathological changes, including hepatomegaly, granulomas, hepatitis with small foci of necrosis and granulomas (Nunura et al., 2010). Therefore, in our opinion, it is quite relevant to carry out research on the clinical manifestation and biochemical profile of blood serum in dogs for toxoplasmosis, which will help veterinarians and animal owners in timely diagnosis and choosing a treatment protocol for dogs.

The purpose of this study was to determine the characteristics of the clinical manifestation and biochemical indicators of blood serum in dogs positive for toxoplasmosis.

2. Material and Methods

During the research, 78 domestic dogs that reacted positively to toxoplasmosis were divided into groups according to the symptom complex that was established during the first visit to the veterinary clinic and anamnestic data about the animal. Depending on the affected organs or systems, they were divided into five groups: the first group - animals with signs of damage to the neuromuscular system (paresis, paralysis, myositis, behavioral changes, eye damage, epileptic phenomena); the second group - with signs of lesions of the gastrointestinal tract (diarrhea, vomiting); the third group - with signs of lesions of the musculoskeletal system (lameness, arthritis); the fourth group - with signs of skin lesions (itching, atopic dermatitis, alopecia, eczema); the fifth group - with signs of lesions of the genitourinary system (nephritis, urolithiasis). Animals were systematized taking into account age and titer of specific IgG against toxoplasma in blood serum.

Blood serum of seropositive SP (n=37) and seronegative SN (n=35) dogs was used for biochemical studies (AlAT, AsAT, urea, creatinine and glucose). The animals were aged from 3 to 5 years. Whole blood was collected in tubes with a blood coagulation activator (SiO_2), the serum was carefully separated from the formed blood elements no later than 1 hour after blood collection. Blood serum tests were performed on an semi-automatic analyzer Evolution 3000 (Biochemical Systems International S.p.A., Italy) using standard sets of reagents "DAC-SpectroMed s.r.l." (Republic of Moldova). In the blood serum of animals, enzymes were determined: alanine aminotransferase (AlAT, K.F.2.6.1.2) and aspartate aminotransferase (AsAT, K.F.2.6.1.1) according to the Reitman-Frankel method and creatinine according to the Jaffe method, urea according to the color reaction with diacetyl monooxime, glucose - by the glucooxidase method (Vlizlo et al., 2012).

3. Results

Table 1 shows the clinical manifestations of toxoplasmosis in dogs. It was established that there is a certain regularity between the average age of dogs, the titer of specific IgG and the clinical manifestation of toxoplasmosis. Thus, in dogs, positive for toxoplasmosis with clinical signs of lesions to the nervous system, the titer of specific IgG was on average 8.48±0.66, which is the highest indicator in comparison with other groups. It was established that signs of lesions to the nervous system appear in 20.5% of cases of toxoplasmosis. The average age of dogs with a symptom complex of nervous system lesions was 4.2±0.23 years.

The number of dogs with clinical signs of skin lesions was 39.4%, which is the highest indicator, while it should be noted that the titer of specific antibodies in these animals was lower than in other groups and was 1.46±0.17 IU on average. The symptom complex of lesions to the gastrointestinal tract manifests itself in an average of 1.3±0.28 years and with relatively low IgG titers (1.73±0.19). Only 6.4% of cases of toxoplasmosis showed clinical signs of lesions to the genitourinary system, the average age of such animals was 6.4±0.67. Lesions of the musculoskeletal system were found in 18% of toxoplasmosis cases with average IgG titers of 4.79±0.53 ($p<0.001$), most often clinical signs appear at an average age of 4.4±0.41 years.

Table 1. Features of the clinical manifestation of toxoplasmosis in dogs (n=78)

Symptocomplexes of lesions	Indexes			
	Number	Percentage ratio, %	The average value of the IgG titer, IU	Average age, years
Nervous system, including organs of vision	16	20.5	8.48 ± 0.66	4.2 ± 0.23
Gastrointestinal tract	12	15.4	1.73 ± 0.19	1.3 ± 0.28
Musculoskeletal system	14	18.0	4.79 ± 0.53**	4.4 ± 0.41
Skin lesions	31	39.7	1.46 ± 0.17	5.4 ± 0.36
Genitourinary system	5	6.4	1.90 ± 0.12*	6.4 ± 0.67

*** p<0.001; *p<0.05*

Table 2. Biochemical indicators of blood serum in SP and SN for canine toxoplasmosis (n=72)

Biochemical indicators of blood serum		Groups of animals		Physiological limits
		Seropositive (Group 1, n=37)	Seronegative (Group 2, n=35)	
AlAT, Units/l	Average indicators	78.14 ± 5.87	65.22 ± 5.21	10-55
	The percentage of animals with a high content	24 (68.5%)	14 (40.0%)	
AsAT, Unit/l	Average indicators	43.4 ± 6.51	31.8 ± 5.18*	10-25
	The percentage of animals with a high content	32 (91.0%)	23 (62.0%)	
Urea, μmol/l	Average indicators	12.56 ± 1.69	10.07±1.32*	3.8-8.3
	The percentage of animals with a high content	28 (75.7%)	15 (43.0%)	
Creatinine, μmol/l	Average indicators	184.3 ± 23.42	163.7 ± 26.68	35-105
	The percentage of animals with a high content	26 (70.3%)	16 (45.6%)	
Glucose, μmol/l	Average indicators	5.84 ± 1.03	6.08 ± 1.06	4.3-6.1
	The percentage of animals with a high content	8 (21.6%)	15 (43.0%)	

$*p< 0.05$

Biochemical analysis of blood serum (Table 2) of SP shows a significant (p<0.05) increase in the levels of liver enzymes, such as AlAT and AsAT. A high level of the average indicator of these enzymes is also noted in the SN dogs, but at the same time, a high level of AlAT was established in 40% of the studied animals, while in the SP - in 68.5%.

The study of the content of urea and creatinine, as indicators that determine the functional and morphological state of the kidneys, in the serum of SP (Group 1) and SN (Group 2) showed that, on average, these indicators were above physiological limits. At the same time, a high content of urea in the blood serum of SP (Group 1) dogs was 1.87 times more frequent than in SN (Group 2). And the content of creatinine was 1.63 times higher compared to the concentration of glucose in the blood serum. It should be noted that in both groups its concentration was within physiological limits. In the first group, in percentage terms, dogs with a high level of glucose in the blood were less often observed.

Therefore, in first group for toxoplasmosis in dogs, the content of such biochemical indicators as AlAT, AsAT, urea, creatinine is much higher than the physiological limits in second group, and more animals have a high concentration of these indicators in their blood. The concentration of glucose in SP (Group 1) animals is on average within physiological limits and the number of animals with a high indicator is almost 2 times less than in SN (Group 2).

4. Discussion

Recent studies that have been analyzed regarding the clinical toxoplasmosis manifestation indicate that dogs rarely have toxoplasmosis as the primary disease, and in most cases the disease is associated with immunosuppression and lack of vaccination against canine distemper virus (CDV) (Calero-Bernal & Gennari Solange, 2019). In our opinion, this is not a sufficiently correct judgment because it is difficult to understand what is the cause of immunosuppression in the absence of immunosuppressive therapy; what does create the conditions for the manifestation of clinical signs - *Toxoplasma gondii* or CDV. Other factors, such as age, gender, housing conditions, etc., may be the trigger for the development of clinical signs. In our research, we analyzed clinical signs in seropositive dogs and found that in 39.7% of dogs, toxoplasmosis is accompanied by skin lesions, while it should be noted that the titer of specific antibodies against toxoplasmosis in such dogs is minimal and averages 1.46±0.171. Manifestation of lesions of the nervous system in dogs is accompanied by a high titer of antibodies (8.48±0.66). The need for a large number of specific IgG is due to the slow diffusion of antibodies through the hematoencephalic and hemato-ophthalmological barriers, as a result of which favorable conditions are created in brain tissues and eyes for the persistence of toxoplasma and the formation of cysts (Frenkel, 1973). The manifestation of lesions of the gastrointestinal tract in dogs due to toxoplasmosis is observed on average at the age of 1.3±0.28 years. According to other researchers, this manifestation is usually associated with viral agents to which animals are sensitive at a young age (Dubey et al., 2020). Taking into account the certain diversity of clinical manifestations and its dependence on age and the titer of specific IgG, it can be assumed that to assess the impact of *Toxoplasma gondii* on the manifestation of clinical signs, a number of factors

should be taken into account. They include the following: the presence of homeless animals in a certain area or country (antigenic pressure), the duration of clinical course taking into account age and breed and biochemical profile of blood serum. Authors have conducted studies to determine the main clinical biochemical indicators of blood serum in SP dogs for toxoplasmosis in comparison with those that applied to the veterinary clinic but were SN. In SP dogs, biochemical analysis of blood serum shows a significant (p<0.05) increase in the level of liver enzymes, such as AlAT and AsAT. A high level of the average indicator of these enzymes is also noted in the SN dogs, but at the same time, a high level of AlAT was established in 40% of the studied animals, while in the SP - in 68.5%. Other studies also consider toxoplasmosis as a disease that causes changes in the metabolic processes of the liver (Atmaca et al., 2013). The obtained results allow us to state that the biochemical profile, in most cases a high level of enzymes such as AlAT and AsAT, can be a marker for further examinations of that animal for toxoplasmosis.

The results of this study show that in dogs, in most cases, toxoplasmosis is accompanied by skin lesions, with a minimal antigenic pressure, as evidenced by the titer of specific IgG. Considering the fact that different clinical signs are manifested by different titers of specific IgG, it can be assumed that *Toxoplasma gondii* can be the main factor in the manifestation of clinical signs in dogs.

Studies of different scientists also consider toxoplasmosis as a liver disease that causes changes in liver metabolic processes (Atmaca et al., 2013), as it was discovered by authors during biochemical analysis of blood serum, which showd a significant increase in the levels of liver enzymes.

Some authors explain the regularity of urea and creatinine content (in particular the frequency of high urea content in the blood serum of seropositive dogs compared to seronegative dogs and the rarity of dogs with high blood glucose levels among seropositive dogs) by the fact that the parasite consumes glucose during metabolic processes (Shehzad et al., 2022).

5. Conclusion

In this article, authors detected clinical signs, that are associated with the manifestation of toxoplasmosis in dogs in most cases. In 39.7% of cases, the clinical manifestation of toxoplasmosis is associated with skin lesions. For the clinical manifestation of toxoplasmosis, which is accompanied by damage of the nervous system, high antigenic activity of the causative agent of toxoplasmosis is probably necessary. This is evidenced by the high content of the titer of specific antibodies. In addition, the biochemical profile of dogs seropositive for toxoplasmosis was shown. A high content of AlAT and AsAT enzymes in blood serum is a factor that indicates the need to determine the presence of the causative agent of toxoplasmosis. The results obtained by us can help veterinarians to assume toxoplasmosis in the earlier period of clinical signs of the disease and prescribe specific treatment accordingly.

Conflict of interest

The authors declare that there is no conflict of interest.

Ethical Approval

For this study, all applicable international, national, and/or institutional guidelines for the care and use of animals were followed during conduction experiments on animals, including current legislation of Ukraine (Article 26 of the Law of Ukraine 5456-VI dated 16.10.2012 “On the Protection of Animals Cruelty Treatment”) and the “General Ethical Principles of Animal Experiments” adopted by the First National Congress on Bioethics (Kyiv, 2001), international bioethical norms (materials of the IV European Convention on the Protection of Vertebrate Animals Used for Experimental and Other purposes (Strasbourg, 1985)) and Directive 2010/63/EU of the European Parliament and of Council of 22 September 2010 on the protection of animals used for scientific.

References

Al-Malki, E.S. (2021). Toxoplasmosis: Stages of the Protozoan Life Cycle and Risk Assessment in Humans and Animals for an Enhanced Awareness and an Improved Socio-economic Status. *Saudi Journal of Biological Sciences*, *28*(1), 962-969. https://doi.org/10.1016/j.sjbs.2020.11.007

Atmaca, H. T., Gazyagci, A. N., Canpolat, S., & Kul, O. (2013). Hepatic Stellate Cells Increase in *Toxoplasma gondii* Infection in Mice. *Parasites & Vectors*, *6*, 135. https://doi.org/10.1186/1756-3305-6-135

Calero-Bernal, R., & Gennari, S. M. (2019). Clinical Toxoplasmosis in Dogs and Cats: An Update. *Frontiers in Veterinary Science*, *6*, 54. https://doi.org/10.3389/fvets.2019.00054

Dubey, J. P., Murata, F. H. A., Cerqueira-Cézar, C. K., Kwok, O. C. H., Yang, Y., & Su, C. (2020). *Toxoplasma gondii* Infections in Dogs: 2009-2020. *Veterinary Parasitology*, *287*, 109223. https://doi.org/10.1016/j.vetpar.2020.109223

Frenkel, J. K. (2000). Biology of *Toxoplasma gondii*. In P. Ambroise-Thomas, & P. E. Petersen (Eds.), *Congenital Toxoplasmosis*: *Scientific Background, Clinical Management and Control* (pp. 9-25). Springer. https://doi.org/10.1007/978-2-8178-0847-5_2

Lashari, M. H., Ghouri, M. T., Akhtar, M. S., Kamran, Z., Chaudhari, M. S., Ayaz, M., Farooq, A. A., & Sarwar, G. (2018). Hematological and Biochemical Alterations Associated with Toxoplasmosis in Dromedaries

(*Camelus dromedarius*) Habitating in Cholistan Desert of Bahawalpur, Punjab, Pakistan. *Journal of Animal and Plant Sciences*, *28*(4), 1043-1048.

Liu, Q., Wang, Z. D., Huang, S. Y., & Zhu X. Q. (2015). Diagnosis of Toxoplasmosis and Typing of *Toxoplasma gondii*. *Parasites & Vectors*, *8*, 292. https://doi.org/10.1186/s13071-015-0902-6

Nunura, J., Vásquez, T., Endo, S., Salazar, D., Rodriguez, A., Pereyra, S., & Solis, H. (2010). Disseminated Toxoplasmosis in an Immunocompetent Patient from Peruvian Amazon. *Revista do Instituto de Medicina Tropical de Sao Paulo*, *52*(2), 107-110. https://doi.org/10.1590/s0036-46652010000200008

Robert-Gangneux, F., & Dardé, M. L. (2012). Epidemiology of and Diagnostic Strategies for Toxoplasmosis. *Clinical Microbiology Reviews*, *25*(2), 264-296. https://doi.org/10.1128/CMR.05013-11

Shehzad, A., Masud, A., Fatima, T., Khan, F. M., Rehman, S., Effendi, M. H., Suwanti, L. T., Khan, I., Tyasningsih, W., Faisal, S., Abadeen, Z. U., & Bibi, S. (2022). Seroprevalence of *Toxoplasma gondii* and Associated Alterations in Hematology and Serum Biochemistry of One-humped Camels (*Camelus dromedarius*) in Pakistan. *Veterinary World*, *15*(1), 110-118. https://doi.org/10.14202/vetworld.2022.110-118

Tenter, A. M., Heckeroth, A. R., & Weiss, L. M. (2000). *Toxoplasma gondii*: from Animals to Humans. *International Journal for Parasitology*, *30*(12-13), 1217-1258. https://doi.org/10.1016/s0020-7519(00)00124-7

Vlizlo, V. V., Fedoruk, R. S., & Ratych I. B. (2012). *Laboratory research methods in biology, animal husbandry and veterinary medicine: A handbook*. Lviv: Spolom. (in Ukrainian)