Shahid Bahonar University of Kerman
Agricultural Biotechnology Journal
Iranian Biotechnology Society
Print ISSN: 2228-6705 Online ISSN: 2228-6500

The correlation of interleukin-6 as an immunological test with canine occult dirofilariasis

Amal Hasan Atiyah

*Corresponding author. Middle Technical University, Institute of Medical Technology/ Baghdad Prosthesis and Orthosis Production Techniques, Baghdad, Iraq. E-mail: dr_amal_h_atiyah@mtu.edu.iq and amhahy2017@gmail.com

Abstract

Objective

Canine occult dirofilariasis (COD) is a subclinical form of Dirofilaria immitis infection, where dogs carry the parasite without exhibiting clinical symptoms. It is unclear how immune cytokines, especially IL-6 and IL-10, affect the immunological response during COD. IL-6 is pro-inflammatory. This study aimed to investigate the correlation of interleukin-6 as an immunological test during COD.

Materials and methods

A total of 56 male and 44 female dogs (total number 100) ranging in age from 1 to 10 years were studied. Aseptic principles were strictly observed for blood collection and sample collection. To examine the presence of microfilaria larvae, peripheral blood smears were prepared from fresh blood. Of the 100 samples tested, 50 were healthy dogs and were used as a control group, and 50 dogs were naturally infected with Dirofilaria immitis. qPCR was used to examine IL-6 gene expression. In addition, the TBP gene was used as a reference gene. SPSS version 29 software was used to analyze the data.

Results

The mean plasma IL-6 levels for the control group and the group with occult dirofilariasis were 56.8 ± 7.6 pg/mL and 124.5 ± 10.2 pg/mL, respectively. The results of the real-time PCR curves and the agarose gel electrophoresis of the PCR products showed that the IL-6 gene is amplified and expressed in the studied dogs. A 114 bp band was observed for this gene and a 184 bp band was observed for the TBP gene in all samples. The mean fold-change of IL-6 gene for the control group was 1.0 ± 0.2 and for the diseased dogs was 4.2 ± 0.9. In the studied samples, a negative correlation coefficient (r = −0.34) was observed for the Pearson correlation between IL-6 levels and parasite load ($p < 0.05$).

Conclusion

The results of this study showed that both plasma levels and gene expression of IL-6 are increased in dogs with latent dirofilariasis. In addition, this increase caused changes in parasite load and was correlated with them. Therefore, IL-6 can be used as a potential inflammatory marker in the management and clinical investigation of this infection. However, before any action, longitudinal studies with larger samples are needed to confirm this role, while simultaneously benefiting from a broader set of biomarkers in the study.

Keywords: canine occult dirofilariasis (COD), dirofilaria immitis infection, interleukin-6, immunological

Paper Type: Research Paper.

Citation: Atiyah, A. H. (2026). The correlation of interleukin-6 as an immunological test with canine occult dirofilariasis. *Agricultural Biotechnology Journal,* 18(1), 213-226.

Agricultural Biotechnology Journal, 18(1), 213-226. DOI: 10.22103/jab.2025.26351.1804

Received: September 27, 2025. Received in revised form: November 25, 2025.

Accepted: November 26, 2025. Published online: January 15, 2026.

Publisher: Shahid Bahonar University of Kerman & Iranian Biotechnology Society.

Introduction

Canine heartworm disease (CHD) is created by Dirofilaria immitis. This parasite is a member of Dirofilaria immitis that is a roundworm. Mosquitoes transmit this disease (Bocková et al., 2015). This disease affects millions of dogs worldwide, making it a major veterinary health concern (Alsarraf et al., 2021; Fuehrer et al., 2021). Dogs may get the illnesses, develop a chronic asymptomatic infection, and often exhibit only moderate symptoms (Alsarraf et al., 2021). The two main methods for detecting heartworm infection are the detection of microfilariae in the blood, which may yield negative results if microfilariae are not produced or if the definitive host's body can destroy these larvae, or where dogs remain asymptomatic and microfilariae are undetectable, referred to as Canine occult dirofilariasis (COD) (Wysmołek et al., 2022; Lensi et al., 2023). Amicrofilaraemic (occult infections) really presents a challenge that is compounded by the absence of effective surveillance systems for parasitic diseases in animals (Portela et al., 2023). The other diagnostic method involves detecting the parasite's antigen, which is serological tests (e.g., ELISA), with highly specific and sensitive detection of active infection (Simón et al., 2007). A Th2-type immune response is more common in microfilaremic infections (mf+) than in amicrofilaremic infections (mf−), indicating that the Th2-type response may play a part in suppressing circulating microfilariae (Oleaga et al., 2009). If an initial immune response to D. immitis occurs, T cells and various cytokines are activated. This activation plays an important role in the inflammatory component of the disease and is described by the fact that cytokines and iNOS mRNA are expressed in the peripheral blood of dogs with natural microfilarial and amicrofilarial infections (Morchón et al., 2007). Cells that play a key role in immune and inflammatory responses produce proteins called cytokines. These cytokines participate in these processes by interfering with the regulation of other cells (Mohammadabadi et al., 2025b). Because they can activate the immune system to provide effective protection, they can be used in vaccine development. Different cells can produce cytokines. For example, epithelial cells produce the cytokines IL-6 or IL-8 (Khezri et al., 2025). Macrophages produce cytokines involved in the activation and regulation of T helper (Th) lymphocytes in inducing an adaptive immune response. The mode of action of all cytokines is similar. They bind to receptors on the surface of target cells and, through these receptors, affect the activity of the target cells (Mohammadabadi et al., 2025a). There are three criteria for classifying cytokines. These criteria include the cells that produce the cytokines, the cells that the cytokines affect, and the type of activity of the cytokines themselves. Cytokines include several groups, although there is considerable overlap between them. These groups include interleukins (IL), interferons (IFN), tumor necrosis factors (TNF), transforming growth factors (TGF), migration inhibitory factors, and smaller chemokines (Mohammadabadi et al., 2025b). IL-6 is a cytokine that performs various functions and has a broad impact on the immune system, and one of its main functions is to increase the host's capacity to fight infections by supporting the immune system (Rose-John et al., 2017). Studies have shown that IL-6 influences the pathogenesis of D. immitis infections and does so by modulating the immune environment in the host (Smith & Maizels, 2014). Several cytokines, including the immunosuppressive ‘interleukin (IL)-10 & "Th2-type" IL-4, have been found to be predominant in manuscripts of the cytokine milieu in mf+ invesions (O'Connor et al., 2003). Yoon et al. (2014) studied dogs with varying degrees of HWD severity. They assessed the therapeutic effect of adult antifungal drugs on inflammatory and cardiovascular biomarkers. They used CRP, IL-6, D-dimer, and cTnI. It was found that leukemia inhibitory factor (LIF), IL-6, cardiotrophin 1 (CT-1), oncostatin M (OSM), ciliary neurotrophic factor (CNTF), IL-11, cardiotrophin-like cytokine factor 1 (CLCF1), and IL-27 cytokines. These eight cytokines are the main members of the IL-6 family. Later, two other cytokines, IL-35 and IL-39, were added to this family. This is because these two cytokines also share gp130 in their receptor complexes (Wang et al., 2016). The multifunctional cytokines of the IL-6 family have a role in immunological responses as well as early development, hematopoiesis, inflammation, bone metabolism, and the growth of the cardiovascular and neurological systems. The concentration of IL-6 was substantially greater in non-survivors than in survivors, according to research using IL-6 as "a prognostic marker in intensive care" medicine (Abramowsky et al., 1981). Therefore, the current study aimed to find immune modulation tests as markers for canine occult dirofilariasis (COD) in dogs naturally infected with Dirofilaria immitis.

Materials and Methods

Blood sample collection: In this study, 5 ml of blood was collected from the femoral and cephalic veins of dogs referred to veterinary clinics in Baghdad and several other cities in the provinces of Iraq, with the help, guidance, and cooperation of veterinary clinic staff. These breeds included native Iraqi breeds and imported breeds. These breeds included Pit Bull, Great Dane, Schnauzer, Balanlong, German Shepherd, Labrador Retriever, Belgian Malinois, Rottweiler, and Doberman. A total of 56 male and 44 female dogs (total number 100) ranging in age from 1 to 10 years were studied. Aseptic principles were strictly observed for blood collection and sample collection.

Sample preparation and initial tests: To examine the presence of microfilaria larvae, peripheral blood smears were prepared from fresh blood. The samples were then directly evaluated under a microscope (Wężyk et al., 2023). Then, a centrifuge was used at a speed of 5000 rpm for 5 minutes. From this amount, 2 ml of each sample was transferred to sterile, flat-bottomed tubes and stored at -20 °C to be used in the next stages of the experiment. The prepared sera were used to perform serological tests for the detection of D. immitis antigen and the analysis of serum markers including IL-6. Next, the Canine IL-6 ELISA Kit was used according to the manufacturer's instructions to measure the concentration of IL-6. The optical absorbance (OD) was measured at a wavelength of 450 nm and the final value was recorded based on the standard curve. Of the 100 samples tested, 50 were healthy dogs and were used as a control group, and 50 dogs were naturally infected with Dirofilaria immitis. After this step, cytokines were measured. In addition, blood samples were collected from 30 healthy and 30 infected dogs (total 60 samples) to be used later for the evaluation of IL-6 gene expression.

Detection of occult infection: A specific antigen test as well as confirmatory PCR were used to determine the presence or absence of latent Dirofilaria immitis infection. Dogs that did not have microfilariae in their blood but had positive antigen or PCR results were classified as Occult Dirofilariasis.

Mononuclear cell extraction and gene expression analysis: A density gradient was used to isolate peripheral blood mononuclear cells (PBMCs). Then, total RNA extraction, measurement of RNA quantity and purity with NanoDrop, and cDNA synthesis with a reverse transcription kit were performed. qPCR was used to examine IL-6 gene expression. In addition, the TBP gene was used as a reference gene. Specific primers (Table 1) were used to amplify the target and reference genes (Kitanaka et al., 2019).

Table 1. Characteristics of primers used to amplify the IL-6 target gene and the TBP reference gene in the studied dogs

Gene	Primer sequence (5′→3′)	Accession number	Amplicon size (bp)
IL-6	F: CAAGATCCTGGTCCAGATGCTAAAG R: CACTCATCCTGCGACTGCAA	NM_001003301.1	114
TBP	F: ACTGTTGGTGGGTCAGCACAAG R: ATGGTGTGTACGGGAGCCAAG	XM_863452	184

The PCR reaction consisted of several steps. The first step was initial denaturation in one cycle, at 95 °C, for 30 seconds. The second step was performed in forty cycles and included denaturation at 95 °C for 5 seconds and annealing/extension at 60 °C for 30 seconds. The $2^{-\Delta\Delta Ct}$ method was used to calculate relative gene expression.

Statistical analysis: SPSS version 29 software was used to analyze the data. Pearson correlation analysis was also used to examine the relationship between different factors. A statistical significance level of $p < 0.05$ was used to make comparisons and examine their significance.

Results and discussion

The mean plasma IL-6 levels for the control group and the group with occult dirofilariasis were 56.8 ± 7.6 pg/mL and 124.5 ± 10.2 pg/mL, respectively. These results show that this level is significantly increased in the affected group compared to the control group ($p < 0.05$). These results indicate that the systemic inflammatory response is activated in dogs with Dirofilaria immitis even in the occult state. These findings are consistent with the results reported by other researchers on the increase in IL-6 in dogs and its relationship with the inflammatory response (Morchón et al., 2007). The results of the quality assessment of the extracted RNAs at the

A260/A280 wavelength showed that the quality was appropriate and desirable (1.8-1.9). The results of the real-time PCR curves and the agarose gel electrophoresis of the PCR products showed that the IL-6 gene is amplified and expressed in the studied dogs. A 114 bp band was observed for this gene and a 184 bp band was observed for the TBP gene in all samples (Figure 1). These observations indicated that the experiment was performed correctly and the desired fragments were amplified correctly.

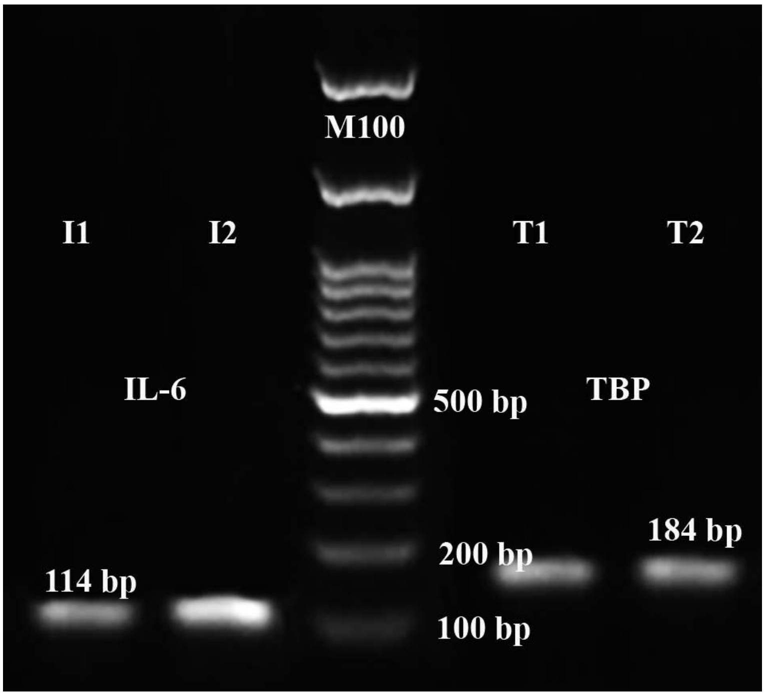

Figure 1. Agarose gel electrophoresis of IL-6 (left) and TBP gene amplification products (right) in the studied dogs. I1 and I2 correspond to the IL-6 gene and T1 and T2 correspond to the TBP gene. M100=size indicator

The mean fold-change of IL-6 gene for the control group was 1.0 ± 0.2 and for the diseased dogs was 4.2 ± 0.9. Comparison between the expression levels of IL-6 gene in the control and diseased groups (Figure 2) shows that IL-6 gene expression in the diseased group is significantly higher than the control group ($p < 0.05$). This observation is probably due to the fact that in the immune response to infection, IL-6 gene is released to play its role at the protein and transcriptional levels. In this regard, Smith et al. (2013) showed that in laboratory models, IL-6 gene plays a key regulatory role in the response to helminth infections and causes host susceptibility to change.

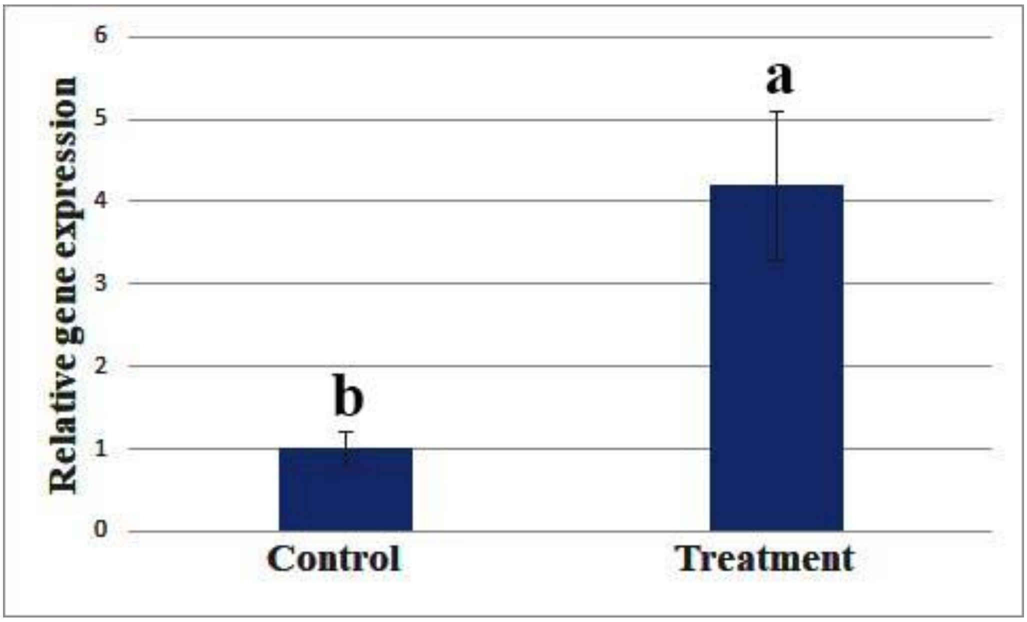

Figure 2. Comparison of IL-6 gene expression levels in control and diseased dogs. Different letters on the bars indicate significant differences between groups (P<0.05)

In the studied samples, a negative correlation coefficient ($r = -0.34$) was observed for the Pearson correlation between IL-6 levels and parasite load ($p < 0.05$). This indicates that an increase in IL-6 may in some cases reduce the parasite load. This means that those who showed a stronger inflammatory immune response (more IL-6) had a lower parasite load. Of course, this negative correlation could also be due to individual differences in immune response, timing of sampling relative to the stage of the parasite cycle, and the presence or absence of microfilariae (occult vs patent). That is, it is possible that in cases where there was a rapid and strong inflammatory response, the parasite load was kept low (Muñoz Caro et al., 2018).

The results observed in latent dirofilariasis indicate that this form of parasitic infection, like other forms, causes elevated serum IL-6 levels and increased expression. The IL-6 gene is a pleiotropic gene, as it is involved in various processes such as T-lymphocyte differentiation, inflammatory responses, and acute phase response. Therefore, its increase is expected when exposed to parasitic antigens (Grebenciucova et al., 2023). Studies on parasite/microbe interactions have shown that D. immitis antigens or microbial co-factors stimulate local immune cells and cause the release of IL-1 and TNF, which in turn alters IL-6 expression (Muñoz Caro, et al., 20018). Several studies have shown that dogs with patent infection have different cytokine expression (e.g. IL-4 and IL-10) compared to occult cases (Morchón et al., 2007). This supports the emergence of cytokine responses in dirofilariasis. Therefore, the results of the present study, which showed that IL-6 expression is increased in occult cases, confirm the results of previous studies on this subject.

In a study by Schüttler & Neumann (2015), clinical studies in hospitalized and critically ill dogs were conducted and showed that high IL-6 levels were associated with worse outcomes and risk of death. They reported that a threshold of 400 pg/mL had high sensitivity and specificity for predicting mortality. In the present study, the mean IL-6 (≈124 pg/mL) in the patient group was reported to be lower than this threshold, but was significantly increased compared to the healthy group. This could indicate a hidden clinical inflammatory response. However, further studies are needed to draw accurate and comprehensive conclusions. Review studies and meta-analyses have shown that in parasitic infections (e.g. malaria) there is often a positive correlation between IL-6 and disease severity. This means that in cases where the disease is more severe, IL-6 levels are higher (Wilairatana et al., 2022). This general pattern confirms the findings of the present article and suggests that IL-6 can be a marker of inflammation and possibly the severity of the immune response.

It should be noted that since the sampling was done at a single point in time, causality between IL-6 and reduction in parasite burden cannot be proven with certainty. This means that longitudinal studies are needed to determine the sequence of cytokine changes and parasite burden. Other factors such as age, race, nutritional status, exposure to antiprotozoa, or concurrent infections should also be controlled for, as these factors can influence IL-6 levels. Although IL-6 measurement can provide valuable information (Goddard et al., 2016), it is better to conduct larger studies to provide a more complete picture. For example, multiple markers such as IL-1, IL-10, and TNF-α should be measured simultaneously with IL-6, along with evaluation of humoral and cellular responses. In addition, IL-6 can be used as an auxiliary biomarker to identify the inflammatory response in dogs with dirofilariasis, even in occult cases. However, before it can be widely used, it is better to establish reference thresholds adapted to the local population and to investigate the sensitivity/specificity of the clinical assay. If a set of cytokines, together with cardiorespiratory parameters and clinical indices, are studied simultaneously with IL-6 in the future, the predictive value of IL-6 at different stages of the disease can be better determined and reported.

Conclusion: The results of this study showed that both plasma levels and gene expression of IL-6 are increased in dogs with latent dirofilariasis. In addition, this increase caused changes in parasite load and was correlated with them. Therefore, IL-6 can be used as a potential inflammatory marker in the management and clinical investigation of this infection. However, before any action, longitudinal studies with larger samples are needed to confirm this role, while simultaneously benefiting from a broader set of biomarkers in the study.

Author contributions

A. A. H.: Conceptualization, methodology, software, validation, formal analysis, investigation, resources, data curation, writing original draft preparation, writing review and editing, visualization, supervision, project administration, funding acquisition. The author has read and agreed to the published version of the manuscript.

Data availability statement

All relevant data supporting the findings of this study are included in the article.

Acknowledgments

The authors are grateful to the Middle Technical University for providing the facilities necessary to conduct this research.

Ethical considerations

This study was approved by the Institute's Scientific-Council of Medical Technology under Administrative Order No. 2/7/27/380B, dated 25/1/2024. It also completed the form designated for obtaining the approval of the Research Ethics Committee, No. 33, authorizing its implementation at the privately owned clinic & the done in the veterinary laboratory services in Al-Khalis District/Diyala. All of the participants were allowed to provide the researcher with the specimens.

Funding

This research did not receive any funding.

Conflict of interest

All authors declared no conflict of interest, financial or otherwise.

References

Abramowsky, C. R., Powers, K. G., Aikawa, M., & Swinehart, G. (1981). *Dirofilaria immitis*: V. Immunopathology of filarial nephropathy in dogs. *American Journal of Pathology, 104*(1), 1–12. https://pubmed.ncbi.nlm.nih.gov/7020425

Alsarraf, M., Levytska, V., Mierzejewska, E. J., Poliukhovych, V., Rodo, A., Alsarraf, M., Kavalevich, D., Dwużnik-Szarek, D., Behnke, J. M., & Bajer, A. (2021). Emerging risk of *Dirofilaria* spp. infection in Northeastern Europe: High prevalence of *Dirofilaria repens* in sled dog kennels from the Baltic countries. *Scientific Reports, 11*(1), Article 1068. https://doi.org/10.1038/s41598-020-80208-1

Bocková, E., Iglódyová, A., & Kočišová, A. (2015). Potential mosquito (Diptera: Culicidae) vectors of *Dirofilaria repens* and *Dirofilaria immitis* in urban areas of eastern Slovakia. *Parasitology Research, 114*(12), 4487–4492. https://doi.org/10.1007/s00436-015-4692-8

Fuehrer, H.-P., Morelli, S., Unterköfler, M. S., Bajer, A., Bakran-Lebl, K., Dwużnik-Szarek, D., Farkas, R., Grandi, G., Heddergott, M., Jokelainen, P., Knific, T., Leschnik, M., Miterpáková, M., Modrý, D., Petersen, H. H., Skírnisson, K., Vergles Rataj, A., Schnyder, M., & Strube, C. (2021). *Dirofilaria* spp. and *Angiostrongylus vasorum*: Current risk of spreading in Central and Northern Europe. *Pathogens, 10*(10), Article 1268. https://doi.org/10.3390/pathogens10101268

Goddard, A., Leisewitz, A. L., Kjelgaard-Hansen, M., Kristensen, A. T., & Schoeman, J. P. (2016). Excessive pro-inflammatory serum cytokine concentrations in virulent canine babesiosis. *PLoS ONE, 11*(3), Article e0150113. https://doi.org/10.1371/journal.pone.0150113

Grebenciucova, E., & VanHaerents, S. (2023). Interleukin 6: At the interface of human health and disease. *Frontiers in Immunology, 14*, Article 1255533. https://doi.org/10.3389/fimmu.2023.1255533

Khezri A, Shafabakhsh H, Alizadeh A, Mohammadabadi M, Shakeri M (2025) Effects of Encapsulated Mixtures of Plant Essential Oils and Organic Acids as an Alternative to Antibiotic Growth Promoters on Humoral Immune Response and Expression of Interleukin-4 and Interferon-Gamma Genes in Broilers. *Journal of Poultry Sciences and Avian Diseases* 3(3): 12-19. https://doi.org/10.61838/kman.jpsad.3.3.3

Lensi, I., Lubas, G., & Papini, R. A. (2023). Incidental finding of *Dirofilaria immitis* (Spirurida: Onchocercidae) microfilariae in the bone marrow of a dog with mixed *Leishmania infantum–Dirofilaria immitis* infection. *Zoonotic Diseases, 3*(2), 162–175. https://doi.org/10.3390/zoonoticdis3020013

Mohammadabadi, M., Khezri, A., Afsharmanesh, M., Kheyrodin, H., Bahreini Behzadi, M. R., Babenko, O., Usenko, S., & Momen, M. (2025a). Investigation of interleukin-6 gene expression in liver and spleen tissues of broiler chickens fed mealworm, probiotics, and mealworm with probiotics. *Agricultural Biotechnology Journal, 17*(4), 469–486. https://doi.org/10.22103/jab.2025.26228.1793

Mohammadabadi, M., Khezri, A., Babenko, O., Borshch, O. O., Kalashnyk, O., Starostenko, I., Tkachenko, S., Klopenko, N., Tytarenko, I., Bezpalyi, I., & Khabiri, A. (2025b). The effect of adding mealworm, probiotics, and mealworm plus probiotics on IL-8 gene expression in liver and spleen tissues of broiler chickens. *OBM Genetics, 9*(4), Article 315. https://doi.org/10.21926/obm.genet.2504315

Morchón, R., López-Belmonte, J., Bazzocchi, C., Grandi, G., Kramer, L. H., & Simón, F. (2007). Dogs with patent *Dirofilaria immitis* infection have higher expression of circulating IL-4, IL-10 and iNOS mRNA than those with occult infection. *Veterinary Immunology and Immunopathology, 115*(1–2), 184–188. https://doi.org/10.1016/j.vetimm.2006.10.019

Muñoz-Caro, T., Conejeros, I., Zhou, E., Pikhovych, A., Gärtner, U., Hermosilla, C., Kulke, D., & Taubert, A. (2018). *Dirofilaria immitis* microfilariae and third-stage larvae induce canine NETosis resulting in different types of neutrophil extracellular traps. *Frontiers in Immunology, 9*, Article 968. https://doi.org/10.3389/fimmu.2018.00968

O'Connor, R. A., Jenson, J. S., Osborne, J., & Devaney, E. (2003). An enduring association? Microfilariae and immunosuppression in lymphatic filariasis. *Trends in Parasitology, 19*(12), 565–570. https://doi.org/10.1016/j.pt.2003.09.017

Oleaga, A., Pérez-Sánchez, R., Pagés, E., Marcos-Atxutegi, C., & Simón, F. (2009). Identification of immunoreactive proteins from the dog heartworm (*Dirofilaria immitis*) differentially recognized by the sera from dogs with patent or occult infections. *Molecular and Biochemical Parasitology, 166*(2), 134–141. https://doi.org/10.1016/j.molbiopara.2009.03.005

Portela, C. S., Mendes de Araújo, C. P., Moura Sousa, P., Gomes Simão, C. L., Silva de Oliveira, J. C., & Crainey, J. L. (2023). Filarial disease in the Brazilian Amazon and emerging opportunities for treatment and control. *Current Research in Parasitology & Vector-Borne Diseases, 5*, Article 100168. https://doi.org/10.1016/j.crpvbd.2023.100168

Rose-John, S., Winthrop, K., & Calabrese, L. (2017). The role of IL-6 in host defence against infections: Immunobiology and clinical implications. *Nature Reviews Rheumatology, 13*(7), 399–409. https://doi.org/10.1038/nrrheum.2017.83

Schüttler, J., & Neumann, S. (2015). Interleukin-6 as a prognostic marker in dogs in an intensive care unit. *Veterinary Clinical Pathology, 44*(2), 223–228. https://doi.org/10.1111/vcp.12247

Simón, F., Kramer, L. H., Román, A., Blasini, W., Morchón, R., Marcos-Atxutegi, C., Grandi, G., & Genchi, C. (2007). Immunopathology of *Dirofilaria immitis* infection. *Veterinary Research Communications, 31*(2), 161–171. https://doi.org/10.1007/s11259-006-3387-0

Smith, K. A., & Maizels, R. M. (2014). IL-6 controls susceptibility to helminth infection by impeding Th2 responsiveness and altering the Treg phenotype in vivo. *European Journal of Immunology, 44*(1), 150–161. https://doi.org/10.1002/eji.201343746

Wang, X., Wei, Y., Xiao, H., Liu, X., Zhang, Y., Han, G., Chen, G., Hou, C., Ma, N., Shen, B., Li, Y., Egwuagu, C. E., & Wang, R. (2016). A novel IL-23p19/Ebi3 (IL-39) cytokine mediates inflammation in lupus-like mice. *European Journal of Immunology, 46*(6), 1343–1350. https://doi.org/10.1002/eji.201546095

Wężyk, D., Romanczuk, K., Rodo, A., Kavalevich, D., & Bajer, A. (2023). Haematological indices and immune response profiles in dogs naturally infected and co-infected with *Dirofilaria repens* and *Babesia canis*. *Scientific Reports, 13*, Article 2028. https://doi.org/10.1038/s41598-023-29011-2

Wilairatana, P., Mala, W., Milanez, G., Kotepui, M., Kotepui, K. U., & Masangkay, F. R. (2022). Increased interleukin-6 levels associated with malaria infection and disease severity: A systematic review and meta-analysis. *Scientific Reports, 12*, Article 5982. https://doi.org/10.1038/s41598-022-09848-9

Wysmołek, M. E., Klockiewicz, M., Długosz, E., & Wiśniewski, M. (2022). Canine antibody response against *Dirofilaria repens* in natural occult and microfilaremic infections. *Comparative Immunology, Microbiology and Infectious Diseases, 86*, Article 101818. https://doi.org/10.1016/j.cimid.2022.101818

Yoon, W. K., Kim, Y. W., Suh, S. L., Choi, R., Lee, S. G., & Hyun, C. (2017). Evaluation of cardiopulmonary and inflammatory markers in dogs with heartworm infection during treatment with the 2014 American Heartworm Society recommended treatment protocol. *Parasites & Vectors, 10*(Suppl 2), Article 535. https://doi.org/10.1186/s13071-017-2427-7

مجله بیوتکنولوژی کشاورزی
شاپا چاپی: ۶۷۰۵-۲۲۲۸ شاپا الکترونیکی: ۶۵۰۰-۲۲۲۸
دانشگاه شهید باهنر کرمان
انجمن بیوتکنولوژی ایران

همبستگی اینترلوکین-٦ به عنوان یک آزمون ایمنی‌شناختی با دیروفیلاریازیس نهفته در سگ (COD)

امل حسن عطیه

نویسنده مسئول: دانشگاه فنی میانه، مؤسسه فناوری پزشکی/ بغداد، رشته فناوری تولید پروتز و ارتز، بغداد، عراق. ایمیل: dr_amal_h_atiyah@mtu.edu.iq و amhahy2017@gmail.com

تاریخ دریافت: ۱۴۰۴/۰۷/۰۵ تاریخ دریافت فایل اصلاح شده نهایی: ۱۴۰۴/۰۹/۰۴ تاریخ پذیرش: ۱۴۰۴/۰۹/۰۵

چکیده

هدف: دیروفیلاریازیس نهفته در سگ (COD) یک شکل تحت‌بالینی از عفونت *Dirofilaria immitis* است که در آن سگ‌ها انگل را بدون نشان دادن علائم بالینی حمل می‌کنند. هنوز مشخص نیست که سیتوکین‌های ایمنی، به‌ویژه IL-6 و IL-10، چگونه بر پاسخ ایمنی در طی COD تأثیر می‌گذارند. اینترلوکین ۶ (IL-6) یک سیتوکین پیش‌التهابی است. هدف این مطالعه بررسی همبستگی اینترلوکین ۶ به عنوان یک آزمون ایمنی‌شناختی در طی COD بود.

مواد و روش‌ها: در مجموع ۵۶ سگ نر و ۴۴ سگ ماده (۱۰۰ حیوان) با دامنه سنی ۱ تا ۱۰ سال مورد بررسی قرار گرفتند. اصول آسپتیک در زمان جمع‌آوری خون و نمونه‌برداری به‌طور دقیق رعایت شد. برای بررسی وجود لاروهای میکروفیلاریا، اسمیر خون محیطی از خون تازه تهیه گردید. از ۱۰۰ نمونه آزمایش‌شده، ۵۰ سگ سالم به عنوان گروه کنترل و ۵۰ سگ به‌طور طبیعی به *Dirofilaria immitis* آلوده بودند. برای بررسی بیان ژن IL-6 از qPCR استفاده شد. همچنین ژن TBP به عنوان ژن مرجع به کار رفت. برای تجزیه و تحلیل داده‌ها از نرم‌افزار SPSS نسخه ۲۹ استفاده شد.

نتایج: میانگین سطح پلاسمایی IL-6 در گروه کنترل و گروه دارای دیروفیلاریازیس نهفته به ترتیب ۷/۶ ± ۵۶/۸ پیکوگرم بر میلی‌لیتر و ۱۰/۲ ± ۱۲۴/۵ پیکوگرم بر میلی‌لیتر بود. نتایج منحنی‌های PCR در زمان واقعی و الکتروفورز ژل آگارز محصولات PCR نشان داد که ژن IL-6 در سگ‌های مورد بررسی تکثیر و بیان می‌شود. یک باند ۱۱۴ جفت‌بازی برای این ژن و یک باند ۱۸۴ جفت‌بازی برای ژن TBP در تمامی نمونه‌ها مشاهده شد. میانگین تغییر بیان (fold-change) ژن IL-6 در گروه کنترل ۰/۲ ± ۱ و در سگ‌های بیمار ۰/۹ ± ۴/۲ بود. در نمونه‌های مورد مطالعه، یک ضریب همبستگی منفی ($r = -0.34$) برای همبستگی پیرسون بین سطوح IL-6 و بار انگل مشاهده شد ($p < 0.05$).

نتیجه‌گیری: نتایج این مطالعه نشان داد که هم سطح پلاسمایی و هم بیان ژنی IL-6 در سگ‌های مبتلا به دیروفیلاریازیس نهفته افزایش می‌یابد. علاوه‌براین، این افزایش باعث تغییر در بار انگل شده و با آن همبستگی داشت. بنابراین، IL-6 می‌تواند به عنوان یک نشانگر التهابی بالقوه در مدیریت و بررسی بالینی این عفونت مورد استفاده قرار گیرد. با این حال، پیش از هرگونه اقدام، مطالعات طولی با حجم نمونه بزرگ‌تر و با استفاده همزمان از مجموعه گسترده‌تری از نشانگرهای زیستی برای تأیید این نقش لازم است.

کلمات کلیدی: ایمنی‌شناسی، اینترلوکین-۶، دیروفیلاریازیس نهفته در سگ (COD)، عفونت *Dirofilaria immitis*

نوع مقاله: پژوهشی

استناد: امل حسن عطیه (۱۴۰۵) همبستگی اینترلوکین-۶ به عنوان یک آزمون ایمنی‌شناختی با دیروفیلاریازیس نهفته در سگ (COD). *مجله بیوتکنولوژی کشاورزی*، ۱۸(۱)، ۲۱۳-۲۲۶.

Publisher: Shahid Bahonar University of Kerman & Iranian Biotechnology Society.