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### Prevalence of *Ehrlichia Canis*, *Borrelia Burgdorferi* and *Dirofilaria Immitis* in Sivas Stray and Shelter Dogs

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#### ABSTRACT

**Objective:** In this study, it was aimed to investigate the prevalence of vector-transportable *Ehrlichia canis*, *Borrelia burgdorferi* and *Dirofilaria immitis* in dogs in Sivas province. **Materials and Methods:** The study was conducted on 100 dogs. Breed, age and sexes of dogs were determined and physical examinations were performed and serum samples were collected for the diagnosis of the diseases. The commercial rapid test kit SNAP\* 4DX\* Plus (IDEXX Laboratories) was used for the detection of *Ehrlichia canis*, *Borrelia burgdorferi* and *Dirofilaria immitis* in dogs. **Results:** In this study, it was determined that 1% of 100 dogs examined were *Ehrlichia canis* and 2% of them were seropositive to *Dirofilaria immitis*, while *Borrelia burgdorferi* seropositive animals were not detected. **Conclusion:** As a result, it was determined that *Ehrlichia canis*, *Borrelia burgdorferi* and *Dirofilaria immitis*, which can be transmitted to dogs by vectors, were observed at low rates in Sivas province. However, at the same time, necessary protective measures should be taken against these diseases that can be transmitted by vectors in this region.

**Keywords:** Borreliosis, *Dirofilaria immitis*, Dog, Ehrlichiosis, Sivas.

### Sivas Sokak ve Barınak Köpeklerinde *Ehrlichia Canis*, *Borrelia Burgdorferi* ve *Dirofilaria Immitis*'in Prevalansı

#### ÖZ

**Amaç:** Bu çalışmada, Sivas ilindeki köpeklerde vektörle taşınabilen *Ehrlichia canis*, *Borrelia burgdorferi* ve *Dirofilaria immitis*'in prevalansının araştırılması amaçlanmıştır. **Gereç ve Yöntem:** Çalışmaya 100 köpek üzerinde yürütülmüştür. Köpeklerin ırk, yaş ve cinsiyetleri belirlenerek fiziksel muayeneleri yapılmıştır ve hastalıkların tanısı için serum örnekleri toplanmıştır. Köpeklerde *Ehrlichia canis*, *Borrelia burgdorferi* ve *Dirofilaria immitis* tespiti için SNAP\* 4DX\* Plus (IDEXX Laboratories) ticari hızlı test kiti kullanılmıştır. **Bulgular:** Bu çalışmada, muayene edilen 100 köpeğin 1 (%1)'inin *Ehrlichia canis* ve 2 (%2)'sinin de *Dirofilaria immitis* yönünden seropozitif olduğu belirlenirken, *Borrelia burgdorferi* yönünden seropozitif hayvan tespit edilmemiştir. **Sonuç:** Sonuç olarak, Sivas ilinde köpeklere vektörlerle taşınabilen *Ehrlichia canis*, *Borrelia burgdorferi* ve *Dirofilaria immitis*'in düşük oranda gözleendiği tespit edilmiştir. Ancak aynı zamanda, bu bölgede vektörlerle taşınabilen bu hastalıklara karşı gerekli koruyucu önlemlerin alınması gerekmektedir.

**Anahtar Kelimeler:** Borreliosis, *Dirofilaria immitis*, Köpek, Ehrlichiosis, Sivas.

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## INTRODUCTION

Vectors and vector-borne diseases have started to gain importance due to the significant climatic and environmental changes occurring today. Vector-borne bacterial, viral, spirochetal and rickettsial diseases with zoonotic importance are transmitted by arthropods (Ataş et al., 1997; İnci and Düzlü, 2009).

Ehrlichiosis (tropical pancytopenia) is a rickettsial disease characterized by decreased formed blood elements and transmitted by ticks to dogs and humans (Eng and Giles, 1989; Matthewman et al., 1993; Friedman et al., 1997).

Lyme disease or Borreliosis; is a zoonotic disease caused by the spirochete *Borrelia burgdorferi*, transmitted mainly by ticks of the genus *Ixodes*, and affecting all organs and systems (Skotarczak and Wodecka, 2003). Lyme disease in dogs was first identified in the United States in the 1980s and has spread worldwide (Koneman et al., 1997).

Dirofilariosis is a parasitic disease transmitted by mosquitoes, especially causing cardiopulmonary diseases. Although the disease usually causes infection in dogs and canines, it rarely occurs in humans and cats (Araujo et al., 2003).

This study aimed to investigate the prevalence of vector-transportable *Ehrlichia canis*, *Borrelia burgdorferi* and *Dirofilaria immitis* in dogs in Sivas province.

## MATERIALS AND METHODS

This study was conducted between April 2017-2018. During this period, a total of 100 dogs, 53 male and 47 female, from different breeds, aged between 10 months and 9 years, weighing between 9-28 kg and living in Sivas animal shelters were used as material.

10 ml of blood was taken from Vena cephalica antebrachium of dogs and put into tubes with and without anticoagulant in equal amounts. Blood samples were taken between 16:00 and 19:00. Blood samples containing anticoagulant (Edta) were examined for microfilaria on the same day. The blood taken into tubes without anticoagulant was centrifuged at 3000 rpm for 8-10 minutes, and their serum was removed and stored at -20 °C until serological testing. This study was carried out with the approval of Sivas Cumhuriyet University Animal Experiments Local Ethics Committee with the letter dated 23.02.2016 and numbered 65202830-050.040.04-26.

### Modified knott method

In the modified knott method, 1 ml of blood sample with anticoagulant was mixed with 9ml of 2% formalin (2ml of 37% concentrated formaldehyde, 100 ml of distilled water) and centrifuged at 2000 rpm for 3.5 minutes. After centrifugation, the liquid on it was poured and 0.1% methylene blue was added as much as the amount of residue remaining at the bottom. Then, a few drops of this mixture were taken and examined in terms of microfilaria under the light

microscope at 10x and 40x magnifications (Wang, 1997).

### Serological method

A commercial rapid test kit (SNAP\* 4Dx\* Plus, IDEXX Laboratories) working with ELISA principle was used for the diagnosis of *Ehrlichia canis*, *Borrelia burgdorferi* and *Dirofilaria immitis* from serum samples. In previous studies with the test kit used, specificity for *Dirofilaria immitis* antigens was 97% (Bowman et al., 2009), sensitivity 84% (Atkins, 2003), specificity for *B. Borrelia burgdorferi* antibodies 99.5% (Duncan et al., 2004), sensitivity 94.4% (O'Connor et al., 2004), specificity for *Ehrlichia canis* antibodies 100% (O'Connor et al. 2004; O'Connor et al., 2006), sensitivity 95.7% (O'Connor et al., 2002) has been determined.

### Statistical analysis

The mean, minimum/maximum values and standard errors of the findings in the study were determined using the SPSS 15.0 computer program.

## RESULTS

In this study, 1 out of 100 dogs (1%) were seropositive for *Ehrlichia canis* and 2 (2%) were seropositive for *Dirofilaria immitis*, but no dogs were found to be seropositive for *Borrelia burgdorferi*.

As a result of the clinical examination, the distribution of the diseases according to the age and sex of the dogs is shown in Table 1-4. While the *Ehrlichia canis* seropositive dog was male, one of the *Dirofilaria immitis* seropositive dogs was male and the other was female. In the clinical examination of the dog determined to be *Ehrlichia canis* seropositive, stagnation, loss of appetite, epistaxis, pallor of the mucous membranes and petechial hemorrhages were determined. It has also been found in dyspnea, exercise intolerance, and fatigue. Disease-related symptoms such as cough, respiratory distress and chest pain were detected in only one of the *Dirofilaria immitis* seropositive dogs.

**Table 1. Distribution of *E. canis* in dogs by sex.**

Gender	Number of Dogs	Number of Infected Dogs	Infection Rate
Male	53	1	1.88
Female	47	0	0
<b>Total</b>	<b>100</b>	<b>1</b>	<b>1</b>

**Table 2. Distribution of *D. Immitis* in dogs by sex.**

Gender	Number of Dogs	Number of Infected Dogs	Infection Rate
Male	53	1	1.88
Female	47	1	2.33
<b>Total</b>	<b>100</b>	<b>2</b>	<b>2</b>

**Table 3. Distribution of *E.canis* in dogs by age.**

Age	Number of Dogs	Number of Infected Dogs	Infection Rate
0-2	39	0	0
3-4	24	1	4
5-6	23	0	0
7-9	14	0	0

**Table 4. Distribution of *D. Immitis* in dogs by age.**

Age	Number of Dogs	Number of Infected Dogs	Infection Rate
0-2	39	0	0
3-4	24	0	0
5-6	23	1	4.34
7-9	14	1	7.14

## DISCUSSION

Vector-borne diseases, which are frequently seen in our country, can cause diseases in both humans and animals and cause significant economic losses in animals (Gazyacı and Aydenizöz 2010; Issi et al., 2010). Canine monocytic ehrlichiosis, which is important in terms of veterinary medicine and spreads in carnivores all over the world, is a disease transmitted to dogs by ticks named *R. sanguineus* caused by *Ehrlichia canis* (Dodurka and Bakirel, 2002; Bremer et al., 2005). In the acute phase of ehrlichiosis, clinical symptoms such as severe weight loss, fever, anorexia, stagnation, ocular and nasal discharge, dyspnea, lymphadenopathy, epistaxis, central nervous system findings, edema in the extremities and scrotum have been reported (Eng and Giles, 1989; Dodurka and Bakirel, 2002). In the chronic phase of the disease, clinical findings such as weakness, depression, anorexia, fever, progressive weight loss, epistaxis, pallor in the mucous membranes and edema, especially in the hind legs and scrotum, can be seen commonly (Smith et al., 1975; Mylonakis et al., 2010; Ural et al., 2014; Waner et al., 2001). Ehrlichiosis in dogs has a wide distribution in the world, being more common in tropical and subtropical regions (Carrade et al., 2011; Unver et al., 2001; Waner et al., 2001). The presence of the disease has been reported in many countries in Asia, Africa, Europe and America (Carrade et al., 2011; Tsachev et al., 2006; Matthewman et al., 1993). Karagenç et al (2005) determined that *Ehrlichia canis* was positive in 41.5% of 371 dogs included in the study from various parts of the Aegean Region such as Manisa, Marmaris, Muğla, Selçuk, Aydın and Bodrum. In a study conducted by Batmaz et al. (2001) in the province of Izmir, they determined that the prevalence of ehrlichiosis was 40.6% with IFAT. Ural et al (2014), on the other hand, reported that the

disease was observed at a rate of 27.5% in a study they conducted on the seroprevalence of ehrlichiosis with the SNAP 4Dx rapid test kit in dogs in the Aegean region. In the present study, 1 out of 100 dogs were determined to be seropositive for *Ehrlichia canis* with the SNAP 4DX Plus test kit. The lower results obtained in this study compared to other studies may be due to the number of samples, the difference in the diagnostic test methods used, and the density of vector ticks that carry the disease. Lyme disease or borreliosis is a spirochete disease caused by *Borrelia burgdorferi*, which can also be seen in humans, dogs, horses, cattle and cats, especially transmitted by ticks of the genus *Ixodes* (Bowman and Nuttall, 2008). In a study conducted in the USA (Little et al., 2014), the prevalence of borreliosis was found to be 7.2%. In the study conducted in Samsun region, it was determined that 10 of 153 dogs were positive by ELISA method (Çakır and Pekmezci, 2020). Borreliosis antibodies were detected by SNAP 4Dx test in only 2 of 307 dogs in Kuşadası and Aydın (Ural et al., 2014). In the present study, borreliosis antibodies could not be detected in 100 dogs with the SNAP 4Dx Plus test in Sivas province. The inability to detect borreliosis antibodies in this study may be related to the fact that the SNAP 4DX test kit used in the diagnosis of the disease can detect borreliosis antibodies only during active infection, the test method used, the number of samples used in the study, the geographical region where the animals live and the ticks carrying the disease are not infected. Dirofilariosis is a zoonotic disease caused by *Dirofilaria immitis*, which is common in many countries of the world (Wang, 1997). Although studies have been carried out in various provinces since 1961, when the infection was first reported in our country (Oytun, 1961), it is difficult to reveal the general distribution of the disease throughout the country. Various test techniques are used in the diagnosis of dirofilariosis. These tests include the use of various antigen test kits (Ural et al., 2014), direct detection of microfilariae in blood, radiography, arteriography, doppler ultrasonography, autopsy or serology methods, which have been increasingly used recently (Wang, 1997). Among these diagnostic methods, serological method; It is widely used today due to its advantages such as ease of use, detection of amicrophilaria infection, high sensitivity and specificity against the disease (Wang, 1997). In this study; Among the test methods reported above, the modified knott and serological method (ELISA, commercial rapid test SNAP 4Dx Plus, IDEXX Laboratories) were used separately for each dog to identify positive dogs. It is stated that sometimes clinical symptoms may not be seen in dogs with dirofilariosis, and in cases of low parasite density, it may generally have a subclinical course (Wang, 1997, Raynaud, 1992). Grieve et al. (1983) stated that clinical symptoms occur when the

number of parasites is above 25 in dirofilariosis, and the infection progresses very severely when the number of parasites is above 100. Right heart failure, which occurs acutely or slowly, can cause clinical symptoms such as exercise intolerance, ascites, and fluid accumulation in the chest and peritoneal cavity in sick animals (Soulsby 1986; Yıldırım, 2004). In the picture of cardiopulmonary disease, clinical symptoms in various forms such as chronic cough, respiratory distress, abnormal lung and heart sounds and arrhythmia occur, and epistaxis may occur due to abnormal blood coagulation in the lung (Yıldırım, 2004). In parallel with the opinions of Yıldırım and Soulsby (Soulsby, 1986; Yıldırım, 2004), exercise intolerance, chronic cough, respiratory distress, abnormal lung and heart sounds were determined in one of the two dogs determined to be seropositive for *Dirofilaria immitis* in this study. No abnormal clinical symptoms were observed in the other dog. This is likely to be explained by the number of parasites, the duration of infection, and the individual susceptibility of the host, as noted by many investigators (Grieve et al., 1983; Raynaud, 1992).

In the present study, although seropositivity was determined in 2 dogs serologically, positive results were obtained in only 1 dog with the modified knott method. This situation, as stated by many researchers (Loehle, 1997; McTier, 1994), in the formation of occult infection despite the absence of microfilaria in the blood; mild infections of animals, amicrofilaremic infections caused by some drugs used for preventive purposes in animals, presence of immature *Dirofilaria* agents in circulation and Dirofilariosis disease caused by single-sex parasites are shown as reasons.

In many studies (Ağaoğlu and Şahin, 1992; Ağaoğlu et al., 2000; Ural et al., 2014; Voyvoda and Paşa, 2004; Yıldırım et al., 2006) on *Dirofilaria immitis* in different regions of Turkey, the presence of the infection has been revealed. In a study conducted around Kayseri, blood samples obtained from 280 dogs were examined for *Dirofilaria immitis* by membrane filtration and ELISA techniques, and a prevalence of 9.6% was determined (Yıldırım et al., 2006). Again, the prevalence of this parasite was found to be 46.2% in Van (Ağaoğlu et al., 2000) and 13.9% in Aydın (Voyvoda and Paşa 2004). Öncel and Vural (2005) investigated the *Dirofilaria immitis* antigen in the blood of 380 stray dogs obtained from Istanbul and Izmir by ELISA and found the seropositivity to be 1.52% in Istanbul, but could not determine seropositivity in Izmir. *Dirofilaria immitis* infection has been found in studies conducted in many different parts of the world. It has been reported to have a prevalence of 12.2% in Russia (Volgina et al., 2013), 0.3-39.7% in the USA (Theis et al., 2001) and 0.6% in Italy (Cringoli et al., 2001). In this study, a serological study was performed using the SNAP 4DX Plus

commercial test kit. The presence of *Dirofilaria immitis* was detected in only 2 (2%) of 100 dogs used as material. The findings obtained in this study are in line with the results of many researchers (Cringoli et al., 2001; Öncel and Vural, 2005; Voyvoda et al., 2004).

## CONCLUSION

In conclusion, it was determined that *Ehrlichia canis*, *Borrelia burgdorferi* and *Dirofilaria immitis*, which can be transmitted to dogs by vectors, were observed at low rates in Sivas province. However, at the same time, necessary protective measures should be taken against these diseases that can be transmitted by vectors in this region. In addition, it was concluded that the low rate of *Ehrlichia canis*, *Borrelia burgdorferi* and *Dirofilaria immitis*, which can be transmitted to dogs by vectors in Sivas province, may be related to the potential vectorin population.

## Conflict of Interest

The authors declare to have no conflicts of interest.

## Author Contributions

**Plan, design:** ZTA; **Material, methods and data collection:** ZTA, OB, UA, AC; **Data analysis and comments:** OB, UA; **Writing and corrections:** ZTA.

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## REFERENCES

- Ağaoğlu, Z., Akgül, Y., Ceylan, E., & Akkan, H. (2000). Van yöresi köpeklerinde *Dirofilaria immitis*' in yaygınlığı. *Yüzüncü Yıl Üniversitesi Veteriner Fakültesi Dergisi*, 11(2), 41-43.
- Ağaoğlu, Z.T., & Şahin, A. (1992). Van'da *Dirofilaria immitis*. *Yüzüncü Yıl Üniversitesi Veteriner Fakültesi Dergisi*, 3, 117-121.
- Araujo, R. T., Marcondes, C. B., Bastos, L. C., & Sartor, D. C. (2003). Canine dirofilariosis in the region of Conceição Lagoon, Florianópolis, and in the Military Police kennel, São José, State of Santa Catarina, Brazil. *Veterinary Parasitology*, 113(3-4), 239-242. [https://doi.org/10.1016/s0304-4017\(03\)00077-3](https://doi.org/10.1016/s0304-4017(03)00077-3)
- Ataş, A. D., Özçelik, S. & Saygı, G. (1997). Sivas sokak köpeklerinde görülen helmint türleri bunların yayılışı ve halk sağlığı yönünden önemi. *Türkiye Parazitoloji Dergisi*, 21(3), 305-309.
- Atkins, C.E., 2003. Comparison of results of three commercial heartworm antigen test kits in dogs with low heartworm burdens. *Journal of the American Veterinary Medical Association*, 222(9), 1221-1223. <https://doi.org/10.2460/javma.2003.222.1221>

- Batmaz, H., Nevo, E., Waner, T., Senturk, S., Yilmaz, Z. & Harri, S. (2001). Seroprevalence of *Ehrlichia canis* antibodies among dogs in Turkey. *Veterinary Record* 148(21), 665-666. <https://doi.org/10.1136/vr.148.21.665>
- Bowman, A. S., & Nuttall, P. A. (Eds.). (2008). *Ticks: biology, disease and control*. Cambridge University Press.
- Bowman, D., Little, S. E., Lorentzen, L., Shields, J., Sullivan, M. P., & Carlin, E. P. (2009). Prevalence and geographic distribution of *Dirofilaria immitis*, *Borrelia burgdorferi*, *Ehrlichia canis*, and *Anaplasma phagocytophilum* in dogs in the United States: Results of a national clinic-based serologic survey. *Veterinary Parasitology*, 160(1-2), 138-148. <https://doi.org/10.1016/j.vetpar.2008.10.093>
- Bremer, W. G., Schaefer, J. J., Wagner, E. R., Ewing, S. A., Rikihisa, Y., Needham, G. R., ... & Stich, R. W. (2005). Transstadial and intrastadial experimental transmission of *Ehrlichia canis* by male *Rhipicephalus sanguineus*. *Veterinary Parasitology*, 131(1-2), 95-105. <https://doi.org/10.1016/j.vetpar.2005.04.030>
- Çakır, K., & Pekmezci, D. (2018). Samsun ili ve çevresindeki köpeklerde Lyme hastalığının serolojik olarak araştırılması. *Journal of Anatolian Environmental and Animal Sciences*, 5(4), 696-703.
- Carrade, D., Foley, J., Sullivan, M., Foley, C. W., & Sykes, J. E. (2011). Spatial distribution of seroprevalence for *Anaplasma phagocytophilum*, *Borrelia burgdorferi*, *Ehrlichia canis*, and *Dirofilaria immitis* in dogs in Washington, Oregon, and California. *Veterinary Clinical Pathology*, 40(3), 293-302. <https://doi.org/10.1111/j.1939-165X.2011.00334.x>
- Cringoli, G., Rinaldi, L., Veneziano, V., & Capelli, G. (2001). A prevalence survey and risk analysis of filariasis in dogs from the Mt. Vesuvius area of southern Italy. *Veterinary Parasitology*, 102(3), 243-252. [https://doi.org/10.1016/S0304-4017\(01\)00529-5](https://doi.org/10.1016/S0304-4017(01)00529-5)
- Dodurka, T., & Bakirel, U. (2002). Bir köpekte Ehrlichiosis olgusu. *Istanbul Üniversitesi Veteriner Fakültesi Dergisi*, 28(1), 11-16.
- Duncan, A. W., Correa, M. T., Levine, J. F., & Breitschwerdt, E. B. (2004). The dog as a sentinel for human infection: prevalence of *Borrelia burgdorferi* C6 antibodies in dogs from southeastern and mid-Atlantic states. *Vector-Borne & Zoonotic Diseases*, 4(3), 221-229. <https://doi.org/10.1089/vbz.2004.4.221>
- Eng, T. R., & Giles, R. (1989). Ehrlichiosis. *Journal of the American Veterinary Medical Association*, 194(4), 497-500.
- Friedman, A. D., Daniel, G. K., & Qureshi, W. A. (1997). Systemic ehrlichiosis presenting as progressive hepatosplenomegaly. *Southern Medical Journal*, 90(6), 656-660. <https://doi.org/10.1097/00007611-199706000-00017>
- Gazyacı, A. N., & Aydenizöz, M. (2010). Keneler ve kenelerin taşıdığı bazı önemli hastalıklar. *Türkiye Parazitoloji Dergisi*, 34(2), 131-136.
- Grieve, R. B., Lok, J. B., & Glickman, L. T. (1983). Epidemiology of canine heartworm infection. *Epidemiologic Reviews*, 5, 220-246. <https://doi.org/10.1093/oxfordjournals.epirev.a036260>
- İnci, A., & Düzlü, Ö. (2009). Vektörler ve vektörlerle bulaşan hastalıklar. *Erciyes Üniversitesi Veteriner Fakültesi Dergisi*, 6(1), 53-63.
- Issi, M., Gul, Y., & Basbug, O. (2011). The effect of classical theileriosis treatment on thyroid hormone levels in cattle naturally infected with *Theileria annulata*. *Asian Journal of Animal and Veterinary Advances*, 6(5), 531-536.
- Karagöç, T., Hoşgör, M., Bilgiç, H., Paşa, S., Kırılı, G., & Eren, H. (2005). Ege bölgesinde köpeklerde *E. canis*, *A. phagocytophila* ve *A. platys*' in prevalansının Nested-PCR ile tespiti. In *Proceedings of the XIV National Parasitology Congress* (pp. 18-25).
- Koneman, E. W. (1997). The Gram-positive cocci. Part II: streptococci, enterococci, and streptococcus-like bacteria. *Color atlas and textbook of diagnostic microbiology*, 577-649.
- Little, S. E., Beall, M. J., Bowman, D. D., Chandrashekar, R., & Stamaris, J. (2014). Canine infection with *Dirofilaria immitis*, *Borrelia burgdorferi*, *Anaplasma spp.*, and *Ehrlichia spp.* in the United States, 2010–2012. *Parasites & Vectors*, 7(1), 1-9. <https://doi.org/10.1186/s13071-020-04514-3>
- Loehle, C. (1997). The pathogen transmission avoidance theory of sexual selection. *Ecological Modelling*, 103(2-3), 231-250. [https://doi.org/10.1016/S0304-3800\(97\)00106-3](https://doi.org/10.1016/S0304-3800(97)00106-3)
- Matthewman, L. A., Kelly, P. J., Bobade, P. A., Tagwira, M., Mason, P. R., Majok, A., ... & Raoult, D. (1993). Infections with *Babesia canis* and *Ehrlichia canis* in dogs in Zimbabwe. *The Veterinary Record*, 133(14), 344-346. <https://doi.org/10.1136/vr.133.14.344>
- McTier, T. L. (1994). A guide to selecting adult heartworm antigen test kits. *Veterinari Medicina* 6,528-543.
- Mylonakis, M. E., Ceron, J. J., Leontides, L., Siarkou, V. I., Martinez, S., Tvarijonavičiute, A., ... & Harrus, S. (2011). Serum acute phase proteins as clinical phase indicators and outcome predictors in naturally occurring canine monocytic ehrlichiosis. *Journal of Veterinary Internal Medicine*, 25(4), 811-817. <https://doi.org/10.1111/j.1939-1676.2011.0728.x>
- O'Connor, T.P., Esty, K.J., Machenry, P., Hanscom, J.L., Bartol, B.A., Lawton, T., 2002. Performance evaluation of *Ehrlichia canis* and *Borrelia burgdorferi* peptides in a new *Dirofilaria immitis* combination assay. In: American Heartworm Society Triannual Symposium. pp. 77–84.
- O'Connor, T. P., Esty, K. J., Hanscom, J. L., Shields, P., & Philipp, M. T. (2004). Dogs vaccinated with common Lyme disease vaccines do not respond to IR6, the conserved immunodominant region of the VlsE surface protein of *Borrelia burgdorferi*. *Clinical and Vaccine Immunology*, 11(3), 458-462. <https://doi.org/10.1128/CDLI.11.3.458-462.2004>

- O'Connor, T. P., Hanscom, J. L., Hegarty, B. C., Groat, R. G., & Breitschwerdt, E. B. (2006). Comparison of an indirect immunofluorescence assay, western blot analysis, and a commercially available ELISA for detection of *Ehrlichia canis* antibodies in canine sera. *American Journal of Veterinary Research*, 67(2), 206-210. <https://doi.org/10.2460/ajvr.67.2.206>
- Öncel, T., & Vural, G. (2005). Seroprevalence of *Dirofilaria immitis* in stray dogs in Istanbul and Izmir. *Turkish Journal of Veterinary and Animal Sciences*, 29(3), 785-789.
- Oytun, H. Ş. (1961). *Genel Parazitoloji ve Helmintoloji*. Ankara Üniversitesi Veteriner Fakültesi Yayınları.
- Raynaud, J. P. (1992). Thiacetarsamide (adulticide) versus melarsomine (RM 340) developed as macrofilaricide (adulticide and larvicide) to cure canine heartworm infection in dogs. *Annales de Recherches Vétérinaires* 23, (1), 1-25.
- Skotarczak, B., & Wodecka, B. (2003). Molecular evidence of the presence of *Borrelia burgdorferi* sensu lato in blood samples taken from dogs in Poland. *Annals of Agricultural and Environmental Medicine*, 10(1), 113-5.
- Smith, R. D., Ristic, M., Huxsoll, D. L., & Baylor, R. A. (1975). Platelet kinetics in canine ehrlichiosis: evidence for increased platelet destruction as the cause of thrombocytopenia. *Infection and Immunity*, 11(6), 1216-1221. <https://doi.org/10.1128/iai.11.6.1216-1221.1975>
- Soulsby, E. J. L. (1986). *Helminths, Protozoa and Arthropods of Domesticated Animal* (7th Edition). Bailliere Tindall.
- Theis, J. H., Stevens, F., Theodoropoulos, G., & Ziedins, A. C. (1999). Studies on the prevalence and distribution of filariasis in dogs from Los Angeles County, California (1996-1998). *Canine Practice*, 24, 8-16.
- Tsachev, I., Kontos, V., Zarkov, I. & Krastev S. (2006). Survey of antibodies reactive with *Ehrlichia canis* among dogs in South Bulgaria. *Revue Médecine Vétérinaire*, 157(10), 481-485.
- Unver, A., Huang, H., & Rikihisa, Y. (2006). Cytokine gene expression by peripheral blood leukocytes in dogs experimentally infected with a new virulent strain of *Ehrlichia canis*. *Annals of the New York Academy of Sciences*, 1078(1), 482-486. <https://doi.org/10.1196/annals.1374.090>
- Ural, K., Gultekin, M., Atasoy, A., & Ulutas, B. (2014). Spatial distribution of vector borne disease agents in dogs in Aegean region, Turkey. *Revista MVZ Córdoba*, 19(2), 4086-4098.
- Volgina, N. S., Romashov, B. V., Romashova, N. B., & Shtannikov, A. V. (2013). Prevalence of borreliosis, anaplasmosis, ehrlichiosis and *Dirofilaria immitis* in dogs and vectors in Voronezh Reserve (Russia). *Comparative Immunology, Microbiology and Infectious Diseases*, 36(6), 567-574. <https://doi.org/10.1016/j.cimid.2013.08.003>
- Voyvoda, H., Pasa, S., Töz, S. Ö., Özbek, Y., & Ertabaklar, H. (2004). Aydın'ın bazı ilçe ve köyleri ile İzmir'in Selçuk ilçesindeki köpeklerde Leishmaniosis ve Dirofilariosis' in prevalansı. *Turkish Journal of Veterinary & Animal Sciences*, 28(6), 1105-1111.
- Waner, T., Harrus, S., Jongejan, F., Bark, H., Keysary, A., & Cornelissen, A. W. (2001). Significance of serological testing for ehrlichial diseases in dogs with special emphasis on the diagnosis of canine monocytic ehrlichiosis caused by *Ehrlichia canis*. *Veterinary Parasitology*, 95(1), 1-15. [https://doi.org/10.1016/s0304-4017\(00\)00407-6](https://doi.org/10.1016/s0304-4017(00)00407-6)
- Wang, L. C. (1997). Canine filarial infections in north Taiwan. *Acta Tropica*, 68(1), 115-120. [https://doi.org/10.1016/s0001-706x\(97\)00081-8](https://doi.org/10.1016/s0001-706x(97)00081-8)
- Yıldırım, A., (2004). Ankara ve çevresindeki köpeklerde filarial etkenlerin prevalansı. *Ankara Üniversitesi Veteriner Fakültesi Dergisi* 51(1), 35-40.
- Yıldırım, A., Ica, A., Atalay, O., Duzlu, O., Inci, A. (2006). Prevalence and epidemiological aspects of *Dirofilaria immitis* in dogs from Kayseri province, Turkey. *Res Vet Sci* 2006; 82: 358-363. <https://doi.org/10.1016/j.rvsc.2006.08.006>