

# Vector-borne diseases imported to Poland between 2021 and 2023

**Key Words** : Animal movements, Dogs, Vector-borne diseases

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## Introduction

Vector-borne diseases are a significant and growing problem in both human and veterinary medicine. The emergence of these diseases is driven by factors such as climate change, deforestation, land-use change, urbanization, population growth, migration, habitat fragmentation, animal movements, and biodiversity loss. In Poland, vector-borne diseases like babesiosis (caused by *Babesia canis*), granulocytic anaplasmosis, and Lyme disease are relatively common in dogs. In recent years, there have also been isolated cases of infections with *Babesia gibsoni*, *Hepatozoon canis*, *Leishmania infantum*, and *Dirofilaria immitis*. The rise in these infections may be due to increased travel with pet dogs to countries where these diseases are prevalent and the influx of Ukrainian war refugees and their animals from endemic areas.

## Objective

The aim of the study was to monitor the occurrence of selected vector-borne diseases in anemic dogs arriving in or returning to Poland from areas endemic for these diseases.

## Materials and Methods

The study, conducted from 2021 to 2023, involved 497 anemic dogs. Among them, 184 were brought to Poland by owners fleeing the war in Ukraine, while the remaining 313 dogs returned from holidays in countries such as Croatia, Turkey, Italy, Bulgaria, Albania, and Romania. Additionally, information was gathered on any anti-ectoparasite prophylaxis that had been sought for the animals (Table 1).

The dogs exhibited various clinical symptoms, including apathy, fever, and pale mucous membranes. Hematological examinations confirmed anemia in all dogs and thrombocytopenia in 413 of them. All blood samples were analyzed using a BIONOTE Vcheck M10 analyzer, which amplifies and detects the DNA of *Leishmania* spp., *Babesia* spp., *Mycoplasma haemocanis*, *Hepatozoon* spp., *Ehrlichia* spp., *Anaplasma* spp., *Rickettsia rickettsii*, and *Bartonella* spp. via real-time PCR (Canine Vector 8 Panel). For comparison, All positive DNA samples were also amplified using standard real-time PCR.

**Table 1.** Non-indigenous and travelling dogs screened for vector-borne diseases, by country of origin or destination

Country	Number of examined dogs	Number of dogs protected against ectoparasites	Number of infected dogs	Number of infected dogs protected against ectoparasites
Ukraine	184	36	39	4
Croatia	96	72	7	2
Turkey	79	65	16	3
Italy	48	42	6	1
Bulgaria	42	30	4	1
Albania	36	31	4	0
Romania	12	8	3	0
Total	497	284	79	11

## Results

Molecular tests revealed the presence of vector-borne pathogens in 79 dogs. **All positive results obtained in the BIONOTE Vcheck M10 analyser were confirmed in a standard real-time PCR test and no false-positive results were found.**

The most diagnosed infection was caused by *Babesia canis* (27 dogs), followed by infections with *Anaplasma phagocytophilum* (20 dogs), *Mycoplasma haemocanis* (15 dogs), *Bartonella henselae* (7 dogs), *Ehrlichia canis* (4 dogs), *Hepatozoon canis* (3 dogs), *Babesia gibsoni* (2 dogs) and *Leishmania infantum* (1 dog).

Most of the sick dogs (n = 39) came from Ukraine. In dogs spending holidays with their owners outside Poland, vector-borne diseases were most often detected after their return from Turkey (n = 16), and next in descending order from Croatia (n = 7), Italy (n = 6), Albania (n = 4), Bulgaria (n = 4) and Romania (n = 3) (Table 2).

The effectiveness of ectoparasite prevention was verified using the F test. For all countries except Ukraine, the null hypothesis (H0) was rejected (confidence intervals  $\neq$  1), indicating statistically significant differences between the groups (Table 3).

## Discussion

The study underscores that the increased frequency of tourist trips with pet dogs to countries where vector-borne diseases are endemic, coupled with the influx of immigrants and their dogs from Ukraine to Poland, heightens the risk of a broader spectrum of diseases, some of which are considered exotic in Poland. Infections like *Babesia canis* or *Anaplasma phagocytophilum* are prevalent among native dogs in Poland and generally do not present significant diagnostic or treatment challenges for veterinarians. However, diseases such as ehrlichiosis, hepatozoonosis, leishmaniasis, haemotropic mycoplasmosis, bartonellosis, and babesiosis caused by *Babesia gibsoni* can pose more complex diagnostic and therapeutic dilemmas in the country. Moreover, distinguishing whether clinical symptoms are attributable to vector-borne diseases can be challenging due to overlapping symptoms with other conditions. Therefore, a definitive diagnosis of vector-borne diseases necessitates more sensitive diagnostic methods.

A significant factor contributing to the prevalence of vector-borne diseases is the inadequate use of protective measures against ectoparasites. Only 13.9% of infected dogs had received ectoparasite prevention, while the majority had not been protected. These findings underscore the importance of preventive measures. It is imperative for veterinarians to consistently educate pet owners about these measures, emphasizing their necessity year-round and especially when pets travel internationally. Such proactive steps can substantially reduce the risk of vector-borne diseases in dogs.

## Conclusion

The broader migration crisis and the rising trend of owners traveling with their dogs to regions endemic with infectious and parasitic diseases are significant risk factors for the emergence of these diseases in Poland. Therefore, implementing preventive measures against ectoparasites and maintaining continuous monitoring of vector-borne diseases, particularly in dogs returning from vacations abroad, are crucial for early detection and timely initiation of appropriate therapy.

**Table 2.** Results of vector-borne disease pathogen isolation and amplification from samples of non-indigenous and travelling dogs by country of origin or destination

Country	Number of dogs infected with the particular pathogen								Total
	<i>Babesia canis</i>	<i>Babesia gibsoni</i>	<i>Anaplasma phagocytophilum</i>	<i>Ehrlichia canis</i>	<i>Hepatozoon canis</i>	<i>Leishmania infantum</i>	<i>Mycoplasma haemocanis</i>	<i>Bartonella henselae</i>	
Ukraine	15	0	12	1	0	0	8	3	39
Croatia	6	0	0	1	0	0	0	0	7
Turkey	0	0	4	0	3	1	4	4	16
Italy	1	0	2	2	0	0	1	0	6
Bulgaria	2	0	2	0	0	0	0	0	4
Albania	1	1	0	0	0	0	2	0	4
Romania	2	1	0	0	0	0	0	0	3
Total	27	2	20	4	3	1	15	7	79

**Table 3.** The effectiveness of ectoparasite prevention and probability of infection in the group of protected non-indigenous and travelling dogs

Country	Number of protected and infected dogs	Number of non-protected and infected dogs	Number of protected and non-infected dogs	Number of non-protected and non-infected dogs	P-value	95 % confidence interval	Odds ratio
Ukraine	4	35	32	113	0.115	0.097–1.260	0.405
Croatia	2	5	70	19	0.009	0.009–0.747	0.111
Turkey	3	13	62	1	$2.845 \times 10^{-11}$	$9.562 \times 10^{-5}$ – $4.478 \times 10^{-2}$	0.005
Italy	1	5	41	1	$2.062 \times 10^{-5}$	$1.123 \times 10^{-4}$ – $1.262 \times 10^{-1}$	0.007
Bulgaria, Romania, Albania	1	10	61	18	$2.007 \times 10^{-5}$	$6.752 \times 10^{-4}$ – $2.415 \times 10^{-1}$	0.031
Total	11	68	266	152	$2.200 \times 10^{-16}$	0.042–0.184	0.092

**This study emphasizes the importance of using the Vcheck M CV8 for screening all 8 pathogens linked to vector-borne diseases, showcasing its effectiveness as a reliable point-of-care PCR tool.**

# Prevalence of *Hepatozoon canis* infection in dogs from the area of Lublin Voivodship

**Key Words :** *Hepatozoon canis*, Dogs, PCR, Poland, Vector-borne diseases

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## Introduction

Canine hepatozoonosis is a tick-borne protozoal disease caused by two species: *H. americanum* and *H. canis*. *H. canis* infections are widespread. In contrast, *H. americanum* infections are exclusive to North America.

Transmission of *Hepatozoon* occurs when dogs ingest the oocysts of the parasite from the definitive host, *R. sanguineus*. Predation of infected vertebrates also serves as a possible route for *H. canis* infection, along with vertical transmission.

Recently, clinical cases of *H. canis* infection in dogs were reported for the first time in Poland. The source of infection, however, remains unidentified. This raises questions about whether these infections were isolated incidents or if *H. canis* infections in Polish dogs are more common but underdiagnosed. To clarify this, molecular monitoring for *H. canis* infections is essential.

## Objective

The aim of the study discussed was to detect *Hepatozoon* genetic material in blood samples from dogs suspected of tick-borne diseases, shedding light on the prevalence and clinical implications of these infections.

## Materials and Methods

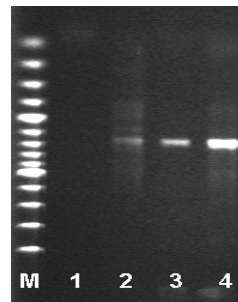
Blood samples were obtained from 107 dogs diagnosed with anemia. Among these, 99 samples were collected from dogs that had never left Polish territory (group 1), while 8 samples were from dogs that had traveled with their owners to Turkey for holidays (group 2).

Using Bionote Vcheck M analyzer, the samples were subjected to real-time PCR to detect DNA from various pathogens, including *Leishmania* spp., *Babesia* spp., *Mycoplasma haemocanis*, *Hepatozoon* spp., *Ehrlichia* spp., *Anaplasma* spp., *Rickettsia rickettsii*, and *Bartonella* spp.. Samples that tested positive for *Hepatozoon* underwent further amplification and sequencing using conventional PCR.

## Results

Using PCR, we detected *Babesia* in 26 samples, *Anaplasma phagocytophilum* in 6 samples, and *H. canis* in 3 samples. Among these, *H. canis* was found in one dog from group 1, which had an infestation of *Ixodes ricinus* ticks, and in two dogs from group 2, which were infested with *Rhipicephalus sanguineus* ticks. These dogs exhibited symptoms such as severe apathy, anorexia, and muscle weakness.

The Vcheck M analyzer provided Ct values ranging from 25 to 28 for these *H. canis* positive samples. Subsequently, conventional PCR and sequencing confirmed the presence of *H. canis* in all three dogs (Figure 1).



**Figure 1.** PCR amplification of a partial sequence of *H. canis*

(M) molecular weight marker = 100 bp; (1) negative control; (2-4) positive samples obtained in own study.

## Conclusion

In Poland, *H. canis* has been sporadically diagnosed, and its primary host, *R. sanguineus* ticks, are only occasionally found in the country. However, These findings emphasize the importance of including *H. canis* in the differential diagnosis of vector-borne diseases, especially in dogs with anemia, in Poland. Moreover, the study underscores the necessity for further research on the roles of different tick species in the transmission of hepatozoonosis. This research is particularly critical given the increasing pet travel and climate changes that could potentially expand the geographic distribution of the disease.

**Due to increased travel and climate change, unusual pathogens may be discovered. This study highlights the need for comprehensive screening tests, such as the Vcheck M CV8, to identify various potential threats.**