# Seroprevalence of Canine Leishmaniasis in Northern Cyprus

# Kuzey Kıbrıs'ta Canine Leishmaniasis'in Prevalansı

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#### **ABSTRACT**

**Objective:** Canine leishmaniasis (CanL) is an important public and veterinary health problem in Mediterranean Basin countries. In this study, we aimed to determine the seroprevalence of CanL in several provinces of Northern Cyprus.

**Methods:** The seroprevalence of CanL was determined by the indirect fluorescent antibody (IFA) test in dog sera. In total, 281 dogs were randomly selected from Nicosia (n=80), Trikomo (n=58), Famagusta (n=60), Morphou (n=30), and Kyrenia (n=53), consistent with a statistically representative number of the regional dog population.

**Results:** Ten (3.55%) out of 281 dogs were found to be seropositive by the IFA test. CanL seropositivity differed between cities as follows: 1.72% (1/58) in Trikomo, 13.20% (7/53) in Kyrenia, 1.67% (1/60) in Famagusta, and 3.33% (1/30) in Morphou. No seropositive dog was found in Nicosia. The symptoms in 37 out of 281 dogs were generalized lymphadenopathy, weight loss, alopecia, exfoliative dermatitis, and epistaxis. Four out of 10 seropositive dogs showed at least one clinical symptom that could be related with CanL.

**Conclusion:** CanL seroprevalence was found to be 3.55% (10/281) in Northern Cyprus. Seropositive dogs, in particular, had lived in areas that exhibited rural as well as urban characteristics.

Keywords: Canine leishmaniasis, IFA test, seroprevalence, Northern Cyprus

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#### ÖZ

Amaç: Canine leishmaniasis (CanL) Akdeniz havzasındaki ülkelerde önemli bir halk sağlığı ve veteriner sağlığı sorunudur. Bu çalışmada, Kuzey Kıbrıs'ın çeşitli illerinde CanL'in seroprevalansının belirlenmesi amaçlandı.

Yöntemler: CanL'nin prevalansı köpek serumlarında Indirekt Floresant Antikor (IFA) testi ile belirlendi. Çalışmada, istatistiksel temsil olarak her bölgedeki toplam köpek sayısı ile uyumlu olmak üzere Lefkoşa (n=80), İskele (n=58), Gazimağusa (n=60), Güzelyurt (n=30) and Girne (n=53) şehirlerinden toplam 281 köpek rastqele seçildi.

**Sonuçlar:** IFA testi uygulanan 281 köpekten 10'unda (3,55%) seropozitiflik saptandı. CanL enfeksiyonuna karşı seropozitiflik oranı şehirler arasında farklılık gösterdi. Seropozitiflik İskele'de %1,72 (1/58), Girne'de %13,20 (7/53), Gazimağusta'da %1,67 (1/60) ve Güzelyurt'da %3,33 (1/30) olarak belirlenirken Lefkoşa'da seropozitiflik saptanmadı. Çalışmadaki köpeklerden 37'sinde generalize lenfadenopati, ağırlık kaybı, alopesi, eksfoliatif dermatit ve burun kanaması belirlendi. Seropozitiflik saptanan 10 köpeğin 4'ünde CanL ile ilişkili klinik semptomlardan en az birinin var olduğu dikkati cekti.

**Sonuç:** Kuzey Kıbrıs'ta CanL seroprevalansı %3,55 (10/281) olarak bulundu. Seropozitifliğin saptandığı 10 köpek kırsal ve kentsel özellikleri birlikte gösteren yörelerde yaşamaktaydı.

Anahtar kelimeler: Canine Leishmaniasis, IFAT, Kuzey Kıbrıs, seroprevalans

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#### INTRODUCTION

Leishmaniasis is a group of parasitic diseases caused by protozoa of the genus Leishmania in mammals. It leads to the development of various pathologies and clinical signs (1-3). Although leishmaniasis is found in many countries in

the Mediterranean Basin (4), there are limited data on the prevalence and distribution of the disease and its impact on public health in Cyprus (5).

Foxes and dogs are the main natural reservoirs for several species of *Leishmania* spp. (6). The emergence of the dis-

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©Copyright 2016 Turkish Society for Parasitology - Available online at www.tparazitolderg.org ©Telif hakkı 2016 Türkiye Parazitoloji Derneği - Makale metnine www.tparazitolderg.org web sayfasından ulaşılabilir. ease in the domestic dog population has become a problem in terms of human health (4, 7-9).

In a sero-epidemiological study conducted in southern Cyprus in 1996, anti-*Leishmania* antibodies were detected in dogs, and the parasite was defined as *L. infantum* zymodem MON 1 (5).

In Mediterranean countries, different from the new world, serological tests form the basis of epidemiological studies on the prevalence of canine leishmaniasis (CanL) (10). The prevalence of CanL differs between countries of the Mediterranean Basin (4). In a two-step study covering Southern Cyprus, the seroprevalence of CanL was found to be 1.7%. A second study was conducted in residential areas intensively inhabited by CanL-seropositive dogs, and the seropositivity for CanL was found to be 10% (5). For the diagnosis of CanL, parasitological, serological, and molecular methods have been used (11).

In this study conducted in Northern Cyprus, we aimed to determine the seroprevalence of CanL using the indirect fluorescent antibody (IFA) test.

#### **METHODS**

This study was conducted between March and June 2007. The numbers of dogs that statistically represent each province were detected, and 281 blood samples were collected from randomly selected dogs of different breeds and genders who were aged from 7 months to 12 years and were living in the cities of Nicosia, Famagusta, Trikomo, Kyrenia, and Morphou, located in Northern Cyprus. The dogs chosen for this study were mainly (98.93%) living outdoors.

Physical examination was performed for each dog, and signs, if any, were noted. In total, 5 mL of venous blood was collected from each dog by brachial vein puncture. The samples were stored at  $+4^{\circ}$ C. Within 5 hours, the samples were centrifuged and sera were stored at  $-20^{\circ}$ C until use.

The antigen was prepared using the *L. infantum* MON 1 strain for the IFA test, and the test was performed as described previously. A twofold serial dilution (1:16 to 1:8192) of dog sera in PBS was used. A titer of <sup>3</sup>1:128 was considered positive for CanL (12, 13).

#### Statistical analysis

Descriptive statistical analyses were performed with SPSS 15.0 (SPSS Inc.; Chicago, IL, USA) packet program and the results were tabulated in Table 1. This study followed the ethical guidelines of animal subjects based on the Declaration of Helsinki.

## **RESULTS**

In this study, the overall seroprevalence of CanL in Northern Cyprus was found to be 3.55% (10/281). The seroprevalence values differed in each city. Kyrenia showed the highest prevalence (13.20%, 7/53), followed by Morphou (3.33%, 1/30), Trikomo (1.72%, 1/58), and Famagusta (1.67%, 1/60). The seroprevalence of CanL according to cities and IFA test dilutions is shown in Table 1.

During clinical examination, 37 out of 281 dogs showed clinical signs such as lymphadenopathy, weight loss, alopecia, exfoliative dermatitis, and epistaxis that could be associated with CanL.

**Table 1.** Distribution of CanL-seropositive dogs according to provinces

			Seropositive dogs	
Provinces	Coordinates	The number of sampled dogs	n	%
Kyrenia	35.338892	53	7	13.2
	33.318705		/	13.2
Morphou	35.198544	30	1	3.3
	32.993746			
Trikomo	35.286201	58	1	1.72
	33.892428			
Famagusta	35.119157	60	1	1.67
	33.932905			
Nicosia	35.201445;	80	0	0
	33.351456			
TOTAL		281	10	3.55

Seropositivity was 10.81% (4/37) in dogs that had one or more clinical symptoms and 2.45% (6/244) in dogs that had no clinical symptoms.

#### **DISCUSSION**

Intense transmission of leishmaniasis by infected sand flies, from dog to dog or from dog to human, occurs in places where the *Leishmania* infection rate is very high in dogs (14). CanL is constantly seen in the entire Mediterranean Coast of Southern European countries, Bosnia and Herzegovina, Croatia, Malta, and Cyprus (15).

In European countries where CanL is endemic, the IFA test was used in 23 of 43 seroprevalence studies, conducted on between 1988 and 1999. The IFA test is considered as the "gold standard" for the serological diagnosis of CanL (10). Although CanL is endemic in countries bordering the Mediterranean Sea, the prevalence of the disease differs between countries and among regions within the same country (4). The prevalence of CanL in different regions of France has been reported to vary between 3.2% and 26.5%. The overall disease prevalence in Greece is 6.8%; it has been determined that this ratio rises to 48.4% in Athens. The prevalence of CanL has been reported to range from 1.4% to 22.2% in Italy and from 19.8% to 5.2% in Spain. The prevalence of CanL is 37.5% and 7% in Algeria and Egypt, respectively; it differs from 10%–21% in Israel and has been reported to be 30.9% in Malta (4, 16-17). The prevalence of CanL ranges from 1.6% to 28.26% in Turkey (18-21). In this study, the seroprevalence of CanL in five different cities was found to range from 1.7% to 13.2% and the overall seroprevalence was found to be 3.55%. These results are similar to those presented in seroprevalence studies previously conducted in countries bordering the Mediterranean Sea.

In this study, the seroprevalence was found to be 13.2% in Kyrenia, 1.72% in Trikomo, 1.67% in Famagusta, and 3.3% in Morphou; however, no seropositivity was observed among 80 samples collected from Nicosia. The rural characteristics of Kyrenia, Trikomo,

Famagusta, and Morphou regions lead to more frequent contact with the vector sand flies of leishmaniasis and dogs. This case explains the higher rates of seropositivity of CanL in these areas in Northern Cyprus. The urban characteristics and geographic features, spraying of insecticides by municipalities, and absence of possible disease vectors in Nicosia should be taken into consideration when discussing the lack of disease prevalence in the area. In a study conducted in Northern Cyprus, the prevalence of CanL among 83 dogs, mainly from Kyrenia and Nicosia provinces, was found to be 3.61% by IFAT (13). In the first phase of the study published in Southern Cyprus, the seroprevalence in randomly selected blood samples collected from 601 dogs was found to be 1.7%. In the second phase of this study, 301 blood samples were taken from residential areas intensively inhabited by CanL-seropositive dogs, and the seroprevalence of CanL in these regions was 10% (5). The present study conducted in Northern Cyprus is similar to the first phase of the study conducted in Southern Cyprus in terms of the method. Overall seroprevalence of CanL in Northern Cyprus was higher than the overall seroprevalence determined in Southern Cyprus. The geographical and demographic characteristics of Northern Cyprus are more suitable for the incidence of the disease than those of Southern Cyprus for the development of CanL. This case may explain the causes of the higher seroprevalence of CanL in Northern Cyprus. A large proportion of dogs with CanL do not show any clinical signs (10, 20, 22). Similar to those findings, in this study, only four out of 10 seropositive dogs (40%) showed one or more clinical symptoms that could be associated with CanL.

Serological analysis of samples showed that the seroprevalence of CanL is 3.55% in Northern Cyprus. CanL is more common in peridomestic areas in Northern Cyprus.

From a geographical point of view, the seroprevalence of CanL in the northern part of Five Finger Mountains is four to seven times higher than that in the southern part. Thus, while planning researches and making disease prevention programs, the geographical distribution of CanL in Northern Cyprus should be taken into consideration.

### CONCLUSION

The seroprevalence of CanL was found to be 3.55% (10/281) in Northern Cyprus.

Seropositive dogs, in particular, had lived in areas that exhibited rural as well as urban characteristics.

**Ethics Committee Approval:** Authors declared that the research was conducted according to the principles of the World Medical Association Declaration of Helsinki "Ethical Principles for Medical Research Involving Human Subjects", (amended in October 2013).

Informed Consent: Not required in this study.

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Hasta Onamı: Bu çalışma için hasta onamına gerek yoktur.

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