Prevalence of *Ctenocephalides canis* and *Ctenocephalides felis* Infestations in Sheep Flocks of Robat Karim, Iran: An Epidemiological Survey

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Abstract

Fleas are considered among the most important arthropods in the fields of veterinary and medical sciences, acting as ectoparasites on various animal species. In certain cases, they also play a role as vectors in the transmission of parasitic zoonotic diseases between animals and humans. These characteristics have made fleas a major concern in global public health and animal health. Sheep, particularly in low- and middle-income countries, are regarded as one of the most important livestock species, playing a significant role in food security and the livelihoods of rural households. Therefore, epidemiological evaluations aimed at identifying factors that adversely affect livestock productivity are of

great importance. This study was conducted to assess the prevalence of infestation with *Ctenocephalides canis* (dog flea) and *Ctenocephalides felis* (cat flea) in sheep populations. A cross-sectional survey was conducted across 19 distinct sheep flocks in Robat Karim County, Tehran Province, Iran. Ectoparasites were collected from 549 sheep (297 male; 252 female). Specimens were transferred to the parasitology laboratory for morphological identification using established diagnostic keys following standard mounting procedures. Overall flea infestation prevalence was 30.6% (168/549 sheep). Among infested animals, *C. canis* and *C. felis* accounted for 77.93% and 22.07% of identified specimens, respectively. Significant geographical variation was observed, with Hesar Mehtar exhibiting the highest infestation rate (63.03%) among surveyed localities. Given fleas' capacity to transmit pathogenic agents and induce anemia in hosts, studies characterizing species-specific prevalence, diversity, and spatiotemporal distribution within defined geographical regions are essential. Such data provide critical foundations for evidence-based interventions, enabling veterinary and public health authorities to implement targeted control strategies and mitigate zoonotic disease risks.

Keywords: *Siphonaptera*, *Ctenocephalides canis*, *Ctenocephalides felis*, Robat Karim, Sheep

1. Introduction

Fleas are small, wingless insects belonging to the order *Siphonaptera* that infest a wide range of mammals and birds worldwide. Due to their hematophagous nature, they are of significant importance in both medical and veterinary fields (1). Some of the most important fleas in these domains belong to the family *Ceratophyllidae*, which includes species such as *Nosopsyllus consimilis* and *Nosopsyllus fasciatus*; the family *Leptopsyllidae*, which includes *Leptopsylla segnis*; and the family *Pulicidae*, which includes *Pulex*

irritans, *Xenopsylla cheopis*, *Ctenocephalides canis*, and *Ctenocephalides felis*. Additionally, fleas act as vectors for zoonotic pathogens, playing a role in the transmission of *Rickettsia felis*, *Bartonella spp.*, *Francisella tularensis*, and *Yersinia pestis* (2)(3)(4).Among them, fleas of the genus *Ctenocephalides* represent a significant group of ectoparasites. *C. canis* and *C. felis* belong to the family *Pulicidae*, order *Siphonaptera*, and class *Insecta* (Table 1).

Table 1. Taxonomy of *Ctenocephalides canis* and *Ctenocephalides felis* parastes

Taxonomic unit	Parasitic group name
Kingdom	Animalia
Phylum	Arthropoda
Class	Insecta
Order	Siphonaptera
Family	Sarcoptidae
Genus	Ctenocephalides
Species	*Ctenocephalides canis Ctenocephalides felis*

They are commonly referred to as the dog flea and cat flea, respectively, and can also function as intermediate hosts for parasitic worms such as *Dipylidium caninum* and *Acanthocheilonema reconditum*. However, they are not strictly host-specific and can parasitize a wide range of mammals, including sheep. Reports indicate the presence of these fleas on various mammals, including sheep, particularly in environments where dogs and cats are in close proximity to livestock. The presence of herding dogs in sheep flocks further facilitates the infestation of *Ctenocephalides* fleas on sheep (5)(6).

However, small ruminants such as sheep do not have a specific flea species exclusively associated with them. Instead, *C. canis* and *C. felis* can infest sheep, leading to blood loss, anemia, itching, and skin hypersensitivity. Iran is a major producer of livestock products, making the control of livestock diseases a

priority. Therefore, it is essential to manage factors that disrupt this industry. In general, the presence of ectoparasites results in reduced livestock productivity, reproductive disorders, skin diseases, and, in severe cases, increased mortality rates. These factors can lead to significant economic losses for farmers and, consequently, negatively impact the livestock industry in the country (7)(8)(9). Given this context, the present study aimed to investigate the prevalence of *C.* fleas in the Robat Karim region in western Tehran Province.

2. Materials and Methods

2.1. Study area

Robat Karim, covering an area of approximately 275 square kilometers, is one of the western regions of Tehran Province. It is located at a longitude of 51°4′, a latitude of 35°28′, and an elevation of 1,100 meters above sea level. The maximum recorded temperature in this region is 44 °C, while the minimum can drop to -20 °C. However, the average annual temperature in Robat Karim is approximately 16 °C. Additionally, the region receives an average annual precipitation of 200 millimeters (Fig. 1).

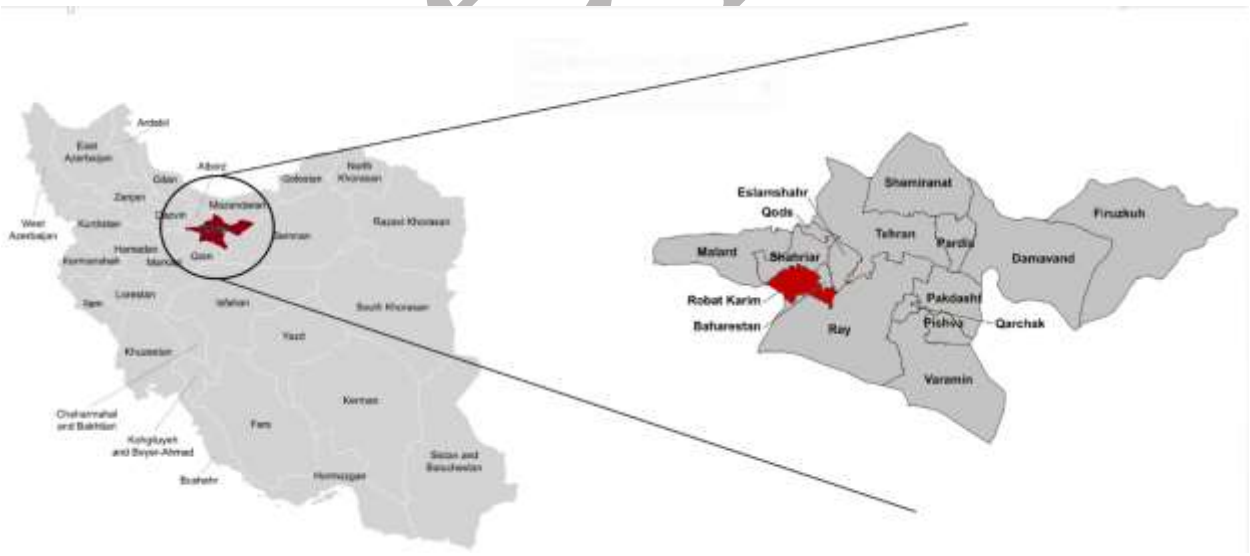

Fig 1. Map of the sampling locations in Robat Karim, Tehran Province, Iran

2.2. Sampling and Preparation of slides

In this cluster-randomized study, sampling was conducted on 549 sheep from 29 different flocks across 10 villages (Peyghambar, Anjemabad, Manjilabad, Shahrabad-e Ilat, Hesar Mehtar, Vahnabad, Hakimabad, Aliabad, Keygavar, Kazemabad) in the Robat Karim region between May 1 and July 31, 2024. All samples were collected between 8:00 AM and 12:00 PM (Table 2). The age of the sheep was determined by interviewing the owners and using dental formula assessment. To collect fleas from the sheep's bodies, an insecticidal spray was first used to immobilize the fleas, followed by combing the animals' fur to capture the fleas. The collected specimens were then placed in 1.5 mL microtubes containing 70% ethanol and transported to the laboratory. For precise identification, the samples were clarified by immersing them in 10% potassium hydroxide (KOH) at room temperature for 24 hours. The fleas were then dehydrated using a graded ethanol series (30%, 50%, 70%, 80%, 90%, and 100%) for 15 minutes at each concentration. Subsequently, the specimens were transferred to ethanol-xylene for one hour, xylene overnight, and lactophenol for 24 hours. After complete mounting, the fleas were examined under a microscope and identified using diagnostic keys (10)(11)

Table 2. Names of villages and number of flocks and sheep studied in the Robat Karim region

The name of the sampling villages	Number of sheep flocks	Number of sheep
Peyghambar	3	75
Anjemabad	4	69
Manjilabad	2	31
Shahrabad-e Ilat	2	40
Hesar Mehtar	2	46
Vahnabad	2	30
Hakimabad	3	53
Aliabad	6	98

Keygavar	3	81
Kazemabad	2	26
10	**29**	**549**

2.3. Statistical analysis

After the final identification of the samples, statistical analysis was performed to determine the relationship between the prevalence of flea infestation and factors such as age, gender, and location of collection. The Chi-square test (2χ) was used for this purpose. In the present study, a significance level of $p \leq 0.05$ was considered, and the statistical analyses were carried out using Microsoft Excel 2016 and 2021 SPSS software.

3. Results

In this study, 549 sheep (297 males and 252 females) were examined to determine the prevalence of *C. canis* and *C. felis* fleas. The results revealed that a total of 168 sheep (94 males and 74 females) were infested with *C. canis* and *C. felis*. The infestation rates, from highest to lowest, were as follows: Hesar Mehtar 63.03%, Shahrabad-e Ilat 37.5%, Vahnabad 36.66%, Peyghambar 36%, Hakimabad 33.96%, Manjilabad 29.03%, Kazemabad 26.92%, Aliabad 23.46%, Anjemabad 20.28%, and Keygavar 18.51%. Additionally, of the 485 fleas collected from the positive samples, 378 (77.93%) were identified as *C. canis* and 107 (22.07%) as *C. felis*. The highest prevalence of *C. canis* was observed in the flocks in the Aliabad region, with a rate of 92.98%, while the highest prevalence of *C. felis* was recorded in Manjilabad at 82.26% (Table 3) and (Figs. 2, 3).

126 **Table 3.** Prevalence of different species of ticks isolated according to the study areas

Name of the area	Number of sheep	Positive case (%)	Negative case (%)	The number of fleas	*Ctenocephalides canis*	*Ctenocephalides felis*
Peyghambar	75	27 (36%)	48 (64%)	85	64 (75.29%)	21 (24.71%)
Anjemabad	69	14 (20.28%)	55 (79.72%)	44	27 (61.36%)	17 (38.64%)
Manjilabad	31	9 (29.03%)	22 (70.97%)	35	6 (17.14%)	29 (82.26%)
Shahrabad-e Ilat	40	15 (37.5%)	25 (62.5%)	43	34 (79.06%)	9 (20.94%)
Hesar Mehtar	46	29 (63.03%)	17 (36.97%)	76	70 (92.10%)	6 (7.9%)
Vahnabad	30	11 (36.66%)	19 (63.34%)	28	25 (89.28%)	3 (10.72%)
Hakimabad	53	18 (33.96%)	35 (66.04%)	43	31 (72.09%)	12 (27.91%)
Aliabad	98	23 (23.46%)	75 (76.54%)	57	53 (92.98%)	4 (7.02%)
Keygavar	81	15 (18.51%)	66 (81.49%)	45	45 (100%)	0 (0%)
Kazemabad	26	7 (26.92%)	19 (73.08%)	29	23 (79.31%)	6 (20.69%)
Total	**549**	**168 (30.6%)**	**381 (69.4%)**	**485**	**378 (77.93%)**	**107 (22.07%)**

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130 **Fig 2-** *Ctenocephalides canis*

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Fig 3- *Ctenocephalides felis*

4. Discussion

This study is the first to examine the prevalence and distribution of *C. canis* and *C. felis* fleas in the Robat Karim region, western Tehran Province. The results revealed an overall flea infestation rate of 30.6%, which is comparable to similar studies on flea prevalence in small ruminants. For instance, a study on external parasites in small ruminants in the Gondar and Banaja regions of Ethiopia reported flea prevalence rates of 35.7% and 36%, respectively .(12)(13) A study conducted in 2023 in the Izmir region of Turkey also found a flea infestation rate of 28.21% in goats (7). Additionally, studies conducted in Iran, specifically in East Azerbaijan in the northwest and in Razavi Khorasan in the northeast, revealed flea prevalence rates of 33% and 65%, respectively, in small ruminants. In both of these studies, the distribution of *C. canis* and *C. felis* was reported as zero, with only *Pulex irritans* being observed (14)(15). Regarding studies that specifically identified flea species, a study in the Tigray region indicated a *C. felis* prevalence of 9% in sheep and 11.15% in goats (16). Furthermore, the prevalence of *C. canis* in Sidi Bouzid, Tunisia, was estimated to be 0.08% (17). In West Azerbaijan, Iran, the prevalence of *C. felis* in sheep was reported as 13%, and in goats, it was 16.8% (18)(19).

The prevalence of *C. canis* and *C. felis* showed no significant relationship with the variables of sex and age. The prevalence rates were very similar between males and females, with the infestation rate in male

sheep estimated at 31.64% and in female sheep at 29.36%. Regarding age, the highest prevalence was found in the >6 years age group (31.35%), while the lowest prevalence was observed in the <1 year age group (28.08%). Furthermore, the highest prevalence of *C. canis* fleas was observed in the 1-3 years age group, while the highest prevalence of *C. felis* was observed in the 3-6 years age group in the Robat Karim region (Table 4). The prevalence of ectoparasites such as fleas depends on various factors, including flock size, hygiene conditions, climate, farming methods, and even the host's immune status (2).

Table 4. Prevalence of different species of ticks isolated according to the study areas

Variable					fleas species	
Sex	**Total**	**N. Infected sheep (%)**	**N. non-infected sheep (%)**	**N. The number of fleas (%)**	**Ctenocephalides canis**	**Ctenocephalides felis**
Male	297	94 (31.64%)	203 (68.36%)	271	222 (81.91%)	49 (18.09%)
Female	252	74 (29.36%)	178 (70.64%)	214	156 (72.89%)	58 (27.10%)
Total	**549**	**168 (30.6%)**	**381 (69.4)**	**485**	**378 (77.93%)**	**107 (22.07%)**
Age grope						
<1	89	25 (28.08%)	64 (71.92%)	81	67 (82.71%)	14 (17.29%)
1-3	243	76 (31.27%)	167 (68.73%)	209	163 (77.99%)	46 (22.01%)
3-6	99	30 (30.3%)	69 (69.7%)	89	60 (67.41%)	29 (32.59%)
>6	118	37 (31.35%)	81 (68.65%)	106	88 (83.01%)	18 (16.99%)
Total	**549**	**168 (30.6%)**	**381 (69.4)**	**485**	**378 (77.93%)**	**107 (22.07%)**

Although in this study, the sex and age variables were not found to be significant, it is essential to consider all aspects in any study. Sheep farming, due to its economic efficiency compared to other animal species, plays a significant financial role in both regional and national economies. Therefore, assessing

the prevalence of diseases in flocks is one of the most important methods for controlling these diseases. Given that fleas are considered one of the most important ectoparasites, we conducted random sampling from 19 sheep farming flocks in 10 villages for this study. However, it should be noted that for more detailed and comprehensive studies, the target population could be expanded.

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The authors declare that all ethical standards have been respected in the conduction of the present research.

Ethical considerations

The authors declare that all ethical standards have been respected in the conduction of the present research.

Conflict of Interest

The authors declare that they have no conflict of interest.

Authors' Contribution

Study concept and design: Al. Gh

Acquisition of data: Al. Gh, Am. Gh, Y. G

Sampling: E. D, Am. Gh, M. H, Al. Gh

Analysis and interpretation of data: M. H, Al. Gh

Drafting of the manuscript: Al. Gh, Am. Gh

Critical revision of the manuscript for important intellectual content: Al. Gh, Y. G

Statistical analysis: Am. Gh, M. H

Administrative, technical, and material support: Al. Gh, Am. Gh, M. H

Data availability

The datasets generated and/or analyzed during the current study are not publicly available due to [e.g., privacy restrictions, institutional policies, or confidentiality agreements], but are available from the corresponding author on reasonable request. Requests will be considered by the corresponding author within the context of ethical and legal standards.

Artificial Intelligence:

This article was written entirely by a human author without the use of AI tools.

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