

Epidemiological studies on sheep and goat *Theileria* infection^{*}

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Summary: This study has been carried out to detect *Theileria* infection in sheep and goats in different geographical regions of Turkey such as Central Anatolia, Eastern Anatolia and Southern Anatolia. Many visits were made to these locations in different periods and a total of 687 sheep and 89 goats, suspected to have *Theileria* infection, were examined. The prevalence of infection with *T. ovis* was 64.19 % in sheep and 12.36 % in goats. Microscopical examination showed that the percentage of parasite carriers was 37.55 % in sheep and 5.62 % in goats. Its seroprevalance was 60.26 % in sheep and 8.99 % in goats by IFAT. *Theileria lestoquardi*, which is very pathogenic for sheep and goats, is not present in Turkey. A total of 709 ticks were collected from sheep and goats.

Key words: Goat, sheep, *Theileria ovis*, *Theileria lestoquardi*

Koyun ve keçilerde *Theileria* enfeksiyonları üzerinde epidemiyolojik araştırmalar

Özet: Bu çalışmada, Orta Anadolu, Güney Anadolu ve Doğu Anadolu gibi Türkiye'nin değişik coğrafik bölgelerinde, koyun ve keçilerde *Theileria* enfeksiyonu incelenmiştir. Değişik zamanlarda bu farklı bölgelerden *Theileria* şüpheli 687 koyun ve 89 keçi muayene edilmiştir. *Theileria ovis*'in hem serolojik, hem de mikroskopik muayenesiyle saptanan prevalans oranları koyunlarda % 64.19 ve keçilerde % 12.36 olarak tespit edilmiştir. Mikroskopik muayenede koyunların % 37.55 ve keçilerin % 5.62'sinde piroplasm şekli görülmüştür. Serolojik muayenede ise IFA testi ile koyunlarda % 60.26 ve keçilerde % 8.99 pozitiflik tespit edilmiştir. Keçi ve koyunlarda yüksek oranda hastalık ve ölüm meydana getirdiği belirtilen patojen *T. lestoquardi* türüne Türkiye'de rastlanmamıştır. Muayene edilen koyun ve keçiler üzerinde toplam 709 adet kene bulunmuştur.

Anahtar sözcükler: Keçi, koyun, *Theileria ovis*, *Theileria lestoquardi*

Introduction

Small ruminant theileriosis develops in erythrocytes in peripheral blood and in lymphoid cells within the reticuloendothelial system. *Theileria lestoquardi* (Syn. *T. hirci*) and the newly described *Theileria* sp. (China 1) cause malignant theileriosis in sheep and goats, a severe disease which ends with death. *Theileria ovis* and *T. separata* cause low or non-pathogenic theileriosis in sheep and goats, which is stated to cause temporary clinical symptoms but not any disease or death (7, 10).

This study was aimed to research the epidemiology of theileriosis in sheep and goats in different geographical regions of Turkey.

Materials and Methods

During the research (September 1997 and March 1999), a total of 687 sheep, 620 of which were from Central Anatolia (453 from Ankara, 85 from Aksaray, 82 from Çankırı), 38 of which were from Eastern Anatolia (21 from Elazığ, 12 from Van, 5 from Bingöl), and 29 of

which were from Southern Anatolia (Mersin) which were over the age of 2 and selected from the herd at random by the method of scanning; and a total of 89 goats, 61 of which were again from Central Anatolia (43 from Ankara, 16 from Çankırı, 2 from Aksaray), and 28 from Southern Anatolia (Mersin) were examined for theileriosis by clinical, microscopical and serological methods (IFAT).

Blood samples collected from the jugular veins of the animals into non-heparinized sterile tubes. Blood smears were made from the punctured ears of the animals. Smears were also made from the materials obtained from lymphnodes by biopsy. The animals were examined for ticks and the ticks which were present were collected. In laboratory, sera were separated from non-heparinized blood and tested for anti-*Theileria* antibody using the Indirect Fluorescent Antibody test. The blood and lymphnode material smears were fixed with methanol, stained with Giemsa and examined for *T. ovis* piroplasms and schizonts, respectively. Besides, the genera and species of the collected ticks were identified.

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Table 1. Prevalence and distribution of *T. ovis* in sheep by using IFAT and microscopical examination.
Tablo 1. Koyunlarda mikroskopik muayene ve IFA testi ile *T. ovis*'in prevalansı ve bölgesel dağılımı

Regions	NSS	M.E		IFAT		M.E + IFAT	
		Positive	%	Positive	%	Positive	%
Central Anatolia	620	239	38.55	392	63.23	412	66.45
Eastern Anatolia	38	7	18.42	17	44.74	17	44.74
Southern Anatolia	29	12	41.38	5	17.24	12	41.38
Total	687	258	37.55	414	60.26	441	64.19

M.E: Microscopical Examination; IFAT: Indirect Fluorescent Antibody Test; NSS: Number of sheep sampled
M.E: Mikroskopik Muayene; IFAT: Indirect Fluorescent Antikor Testi; NSS: Örnek koyun sayısı

Table 2. Prevalence and distribution of *T. ovis* in goats by using IFAT and microscopical examination.
Tablo 2. Keçilerde mikroskopik muayene ve IFA testi ile *T. ovis*'in prevalansı ve bölgesel dağılımı

Regions	NGS	M.E		IFAT		M.E + IFAT	
		Positive	%	Positive	%	Positive	%
Central Anatolia	61	5	8.20	5	8.20	8	13.11
Southern Anatolia	28	0	0	3	10.71	3	10.71
Total	89	5	5.62	8	8.99	11	12.36

M.E: Microscopical Examination; IFAT: Indirect Fluorescent Antibody Test; NGS: Number of goats sampled
M.E: Mikroskopik Muayene; IFAT: Indirect Fluorescent Antikor Testi; NGS: Örnek keçi sayısı

Table 3. Distribution of ticks species collected from sheep and goats in Central Anatolia.
Tablo 3. Orta Anadolu'da koyun ve keçilerden toplanan kene türlerinin dağılımı

Ticks species	Total		Sheep		Goats	
	Tick number	%	Tick number	%	Tick number	%
<i>Rhipicephalus turanicus</i>	285	40.20	172	60.35	113	39.65
<i>Rhipicephalus bursa</i>	221	31.17	49	22.17	172	77.83
<i>Haemaphysalis parva</i>	113	15.94	109	96.46	4	3.54
<i>Haemaphysalis sulcata</i>	44	6.21	43	97.73	1	2.27
<i>Haemaphysalis punctata</i>	1	0.14	1	100.00	0	0.00
<i>Dermacentor marginatus</i>	11	1.55	9	81.82	2	18.18
<i>Dermacentor niveus</i>	4	0.56	0	0.00	4	100.00
<i>Hyalomma marginatum</i>	4	0.56	1	25.00	3	75.00
<i>Ornithodoros lahorensis</i>	26	3.67	26	100.00	0	0.00
Total	709	100.00	410	57.83	299	42.17

For Indirect Fluorescent Antibody (IFA) testing, *Theileria ovis* piroplasm antigen was prepared in the laboratory of the Protozoology and Entomology Department of Ankara University, and *T. lestoquardi* shizont antigen and the positive and negative control serums were provided from Edinburgh University, Centre for Tropical Veterinary Medicine. For *Theileria ovis* piroplasm antigen 1:20, and for *T. lestoquardi* shizont antigen 1:40 and higher dilution steps were accepted to be positive.

Results

In this study, the prevalence of *T. ovis* infection in sheep was determined to be 64.19 %. Microscopical examination of blood smears showed that the percentage of parasite carriers in sheep was 37.55 %. When anti-*Theileria ovis* antigen was used, the seroprevalence of this parasite was 60.26 % seropositive in these regions (Table 1). As also understood from Table 1, *T. ovis*

infection is detected to be more common in sheep in Central Anatolia when compared to the other two regions.

On the other hand, it was seen that in these 2 regions, 12.36 % was positive for the prevalence of *T. ovis* infection in goats. Of these goats 5.62 % carried the parasite and 8.99 % was seropositive for *T. ovis* (Table 2). As also stated in Table 2, the *T. ovis* infection prevalence in goats in Central Anatolia is more common than the ones in Southern Anatolia.

In addition, these animals were detected to be negative in serological and microscopical examinations in terms of *T. lestoquardi*. Parasitaemia level in the blood of the infected sheep was very low (0.05 % to 0.50 %). Microscopical examination of lymphnode biopsy material smears indicated that no schizont forms of the parasite were present in lymphocytes in both the animals. None of the clinical symptoms of theileriosis occurred in the infected sheep and goats. Neither disease nor death cases were present among the infected sheep and goats.

It is stated in Table 3 that in total 709 ticks were collected from goats and sheep in Central Anatolia. 410 (57.83 %) of the 709 ticks were found in sheep and 299 (42.17 %) were found in goats. These were *Rhipicephalus turanicus* (40.20 %), *Rh. bursa* (31.17 %), *Haemaphysalis parva* (15.94 %), *Hae. sulcata* (6.21 %), *Hae. punctata* (0.14 %), *Dermacentor marginatus* (1.55 %), *D. niveus* (0.56 %), *Hyalomma marginatum* (0.56 %) and *Ornithodoros lahorensis* (3.67 %).

Discussion and Conclusion

Lestoquard and Ekrem (8) were the first to state the presence of *T. ovis* in sheep in Turkey. Later, Baumann (2) defined a *Theileria* sp. as *T. hirci* (*T. lestoquardi*) which he had found in Ankara during a postmortem examination. Göksu (3) came across *T. ovis* in Ankara region during the microscopical examination of blood smears of healthy sheep at a rate of 18.26 %. Hoffmann et al. (4) detected *T. ovis* in Turkey during the microscopical examination of the blood smears of sheep and goats at rates of 3.1 % and 16.9 %, respectively. They stated the possibility of the parasite found in goats being *T. hirci*. Recently, in certain cities in Southeastern Anatolia and Malatya, *Theileria* sp. prevalence was detected at a rate of 1- 4 % in sheep and goats (9), and in Kayseri region 18.4 % in sheep and 6 % in goats during microscopical examinations (6).

In the present survey, *Theileria* infection detected in sheep and goats was determined to be more common when compared to the studies realized by other researchers (3, 4, 6, 9). However, previous studies dealt with infections diagnosed by microscopic examination of blood smears and clinical signs of ovine theileriosis, and there was not any serological study to comparatively evaluate the distribution and prevalence of *T. ovis* in Turkey. On the other hand, as also stated by the above mentioned researchers (3, 4), the parasitaemia level of the *Theileria* infected sheep and goat was found to be low and, schizonts were not encountered. Also, no cases of clinical theileriosis or death due to the disease were observed in their surveys (3, 4). These findings show that *Theileria* sp. causing infection in sheep and goats in Turkey is apathogen. In addition, based on the determination of the presence of *Rh. bursa* which is also the carrier of the disease, the *Theileria* sp. found in Turkey was identified as *T. ovis* (3, 4). However, although some researchers (2, 4) did not encounter any disease or death, high rates of parasitaemia in postmortem and alive examinations or schizonts in high numbers, they stated that the *Theileria* species found in Turkey might be *T. hirci*. However, the findings obtained after this study suggested that *T. lestoquardi* was not present in Turkey.

In the same goats and sheep, many *Rh. bursa*, *Rh. turanicus*, *Hae. parva* and *Hae. sulcata*, *D. niveus*, *H. a. anatolicum* ticks species were found. Previous studies (5, 10) show that *Rh. bursa*, *Hae. sulcata* and *H. a. anatolicum* transmit *T. ovis*. Aktaş et al. (1) reported that *Rh. bursa* collected from sheep and goats in Eastern Anatolia were found to be infected with *T. ovis* at a rate of 19.27 % by using PCR and it was emphasized that the *T. ovis* vector might be present in that region. According to our results, *Rh. bursa* was found in sheep and goats in Central Anatolia at a rate of 31.17 %. This indicates that *Rh. bursa* may be the first cause of *T. ovis* infection in this region.

As a result, although *Theileria ovis* was found in the serological and microscopical examinations of sheep and goats in Turkey, *Theileria lestoquardi* was not encountered in this study.

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