

Tick attachment sites in humans living in the Tokat province of Turkey

Tokat ilinde yaşayan insanlardaki kene tutunma bölgelerinin değerlendirilmesi

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ABSTRACT

Objective: The aim of this study was to evaluate the attachment sites of ticks and the demographic properties of patient infested by ticks in Tokat province, a Crimean-Congo hemorrhagic fever (CCHF) endemic region in Turkey.

Methods: In 2009, 5,089 patients with tick bites admitted to hospitals of Tokat province. A standard questionnaire, including the name, age, gender, profession, date, living area, and travel history of patients, was filled by health personals for each patient with a tick bite. Attachment sites of ticks were divided into 9 groups: abdomen, arms, axilla, back, chest, head and neck, hip, legs, and perineum, while the age-group of patients were as follows: 0-9, 10-19, 20-39, 40-64 and ≥65.

Results: The majority of the patients applied (n=1,051, 23.3%) were found ticks on the legs of them. 20-39 year age group tick-infested were the highest proportion (n=1228, 27.24%), while 2,825 (62.67%) of patients were male, and 1,683 (37.33%) were female. In addition, it was determined that 2,740 (60.78%) of patients were living in rural, while 1,768 (39.22%) were living in urban areas. A total of 20 tick taxa were identified, comprising 6 genera: Argas (1 species), Dermacentor (2 species), Haemaphysalis (3 species and

ÖZET

Amaç: Bu çalışmada, Türkiye'de Kırım Kongo Kanamalı Ateşi (KKKA) açısından endemik olan bir bölgede kene enfestasyonu olan kişilerin demografik özelliklerinin ve kenelerin tutunma bölgelerinin değerlendirilmesi amaçlanmıştır.

Yöntem: 2009 yılında Tokat ilinde 5,089 kişi kene enfestasyonu şikayeti ile hastanelere başvurmuştur. Kene tutunması olan her hasta için sağlık personelleri tarafından doldurulan hastaların isim, yaş, cinsiyet, meslek, tarih, yaşadığı alan, seyahat bilgisi ve ek notları içeren standart formlar incelenmiştir. Kene tutunma bölgeleri karın, kollar, koltuk altı, sırt, göğüs, baş ve boyun, kalça, bacaklar ve perine olmak üzere 9 gruba, hastaların yaşları da 0-9, 10-19, 20-39, 40-64 ve ≥65 olmak üzere 5 gruba ayrılarak incelenmiştir.

Bulgular: Başvuran kişilerin büyük bir kısmında (n=1,051, %23,3) kenelerin bacaklara tutunduğu tespit edilmiştir. 20-39 yaş grubundaki kişiler (n=1.228, %27,24) kene tutunması oranının en yüksek olduğu grup olarak belirlenmiştir. Hastalardan 2.825 (%62,67)'inin erkek, 1.683 (%37,33)'inin kadın olduğu tespit edilmiştir. Ayrıca, 2.740 (%60,78) hastanın kırsal alanlarda, 1.768 (%39,22) hastanın kentsel alanlarda yaşadığı belirlenmiştir. İnsanlar üzerinden toplanan kenelerin tür teşhisleri yapıldığında, örneklerin Argas (1 tür), Dermacentor

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1 subspecies), *Hyalomma* (4 species), *Ixodes* (5 species) and *Rhipicephalus* (4 species).

Conclusion: In the present study, attachment sites of ticks and the demographic properties of patients were evaluated for the first time in Tokat province, a CCHF endemic region. In addition, it was reported people infestations by *argas vespertilionis* and *Ixodes gibbosus* ticks for the first time in Turkey.

Key Words: CCHF, human infestation, tick, Tokat, Turkey

(2 tür), *Haemaphysalis* (3 tür ve 1 alttür), *Hyalomma* (4 tür), *Ixodes* (5 tür) ve *Rhipicephalus* (4 tür) cinsleri içerisinde toplam 20 taksona ait olduğu belirlenmiştir.

Sonuç: Bu çalışmayla birlikte, KKKA endemik Tokat bölgesinde kene enfestasyonu olan hastaların demografik özellikleri ve kenelerin tutunma bölgeleri ilk kez değerlendirilmiştir. Ayrıca, Türkiye’de insanların *Argas vespertilionis* ve *Ixodes gibbosus* türü keneler tarafından enfeste edildikleri ilk kez tespit edilmiştir.

Anahtar Kelimeler: KKKA, insan enfestasyonu, kene, Tokat, Türkiye

INTRODUCTION

Ticks are one of the most encountered human parasites throughout the world. They may play an important role in the transmission of several zoonotic diseases to humans, such as Crimean-Congo Hemorrhagic Fever (CCHF), Lyme diseases, rickettsiosis, tick-borne encephalitis (TBE), ehrlichiosis and babesiosis (1-3). Every year thousands of people are affected by tick bites, while hundreds of them are infected with CCHF in Turkey (4-6). According to the Ministry of Health, 9,046 CCHF cases, and 440 deaths were recorded in Turkey between 2002 and 2014. Transmission of CCHF virus generally occurs through tick bites, however the virus can be transmitted by direct contact with blood, tissue or body fluids of patients or viremic animals (7, 8). Mortality rates may reach up to 40% and there is no currently safe and effective CCHF vaccine for humans (9).

In Turkey, CCHF virus has been detected from 7 hard ticks, namely *Haemaphysalis concinna*, *Hyalomma anatolicum*, *Hyalomma marginatum*, *Hyalomma scupence*, *Rhipicephalus bursa*, *Rhipicephalus turanicus* and *Ixodes ricinus* ticks (2), however it is believed that *H. marginatum* is playing a major role in the transmission of CCHF virus (2, 5,

8, 10). The virus has been also detected in several small mammals such as hares and hedgehogs. In addition, antibodies against CCHF virus have been determined in the sera of domestic animals such as horses, donkeys, goats, cattle, sheep, and pigs (11).

Accurate and immediate tick removal from the human skin may be critical because transmission of tick-borne pathogens may occur during the first few hours of the tick attachment (12, 13). For example, *Anaplasma* and *Rickettsia* species are transmitted within 3-6 hours after the tick attachment, whereas *Borrelia burgdorferi* transmission can require 24-48 hours of feeding before a host is infected (14,15). To the best of our knowledge, there is no information regarding the feeding time required for the transmission of CCHF virus from a tick to its host. On the other hand, it is known that certain tick-borne pathogens, such as TBE virus and Powassan virus, are transmitted to the host during the initial minutes of tick attachment or feeding (15, 16). Therefore, knowledge of attachment sites of ticks, duration of tick bites and demographic properties of patients may provide important information for an effective control of ticks and tick-borne diseases (17).

Though many studies on the attachment sites of ticks, duration of tick bites and demographic properties of patients have been conducted (17, 18), there is limited information about these parameters in Turkey (19, 20). The aim of this study was to investigate the species composition of ticks collected from humans in Tokat province in 2009 and to compared the data with previous studies. In addition, attachment sites of ticks and demographic properties of patients should be evaluated for the first time in this CCHF endemic region.

MATERIAL and METHOD

Ticks were collected from humans in Tokat province and its districts in 2009. The ticks were removed from the skin of patients by physicians, nurses, or health technicians (non-attached ticks on the clothes or skin of humans were excluded). Collected ticks were placed in vials with 70% alcohol and sent to the Acarology Laboratory, Department of Biology, Gaziosmanpaşa University (Tokat, Turkey) for identification. Ticks were identified to species based on morphological characteristics using taxonomic keys and descriptions of Filippova (21, 22), Estrada-Peña et al. (23) and Apanaskevich and Horak (24).

A standard questionnaire including name, age, gender, profession, date, living area, and travel history of patients, was filled in for each patient with a tick bite. Questionnaires belonging to 4,508 patients were analyzed statistically, while 581 (11.41%) questionnaires could not be evaluated due to missing data. Informed consent was obtained for all patients.

Chi-square test was used for comparisons of different groups. One variable was tested versus the others in two by two tables. A $p < 0.05$ was considered as statistically significant. Odds ratios are shown with the corresponding 95% confidence intervals. All statistical analyzes were performed using the SPSS software, Version 15.0 (SPSS Inc., USA).

RESULTS

Demographic characteristics of the patients

Out of 4,508 patients 2,825 (62.67%) were male and 1,683 (37.33%) were female. The number of patients living in rural areas was 2,740 (60.78%) while of those living in urban areas was 1,768 (39.22%). Patients living in urban areas were mainly infested in crop fields, gardens, picnic areas or farms. The youngest patient was a one-month-old while the oldest patient was 104-years-old (mean age: 32.71 ± 20.56). The largest proportion of tick bites were seen in 40-64 ($n=1,346$, 29.86%), 20-39 ($n=1,228$, 27.24%) and 10-19 ($n=949$, 21.05%) age groups (Table 1).

Species composition of ticks infesting humans

In 2009, a total of 6,576 ticks were collected from 5,089 patients in Tokat province; however, only 4,508 patients gave accurate information for the questionnaires. A total of 20 tick taxa were identified, comprising 6 genera: *Argas* (1 species), *Dermacentor* (2 species), *Haemaphysalis* (3 species and 1 subspecies), *Hyalomma* (4 species), *Ixodes* (5 species) and *Rhipicephalus* (4 species) (Tables 2 and 3). *Hyalomma marginatum* comprised 70.9% ($n=3,198$) of all ticks removed from patients. Other common ticks infesting humans were *Rhipicephalus bursa* 5.06% ($n=228$), *Haemaphysalis parva* 4.7% ($n=212$) and *Rhipicephalus turanicus* 3.88% ($n=175$) (Table 2).

Attachment sites of ticks

It was shown that *H. marginatum* infests legs 1.45 times (OR: 1.45, CI: 1.23 - 1.69), and *Hyalomma* spp. nymphs infest perineum 2.14 times (OR: 2.14, CI: 1.49 - 3.00) more than other anatomical sites of patients. *Haemaphysalis parva* was mainly detected on head and neck area 6.24 times (OR: 6.24, CI: 4.71 - 8.29), more than other anatomical sites, whereas *Dermacentor marginatus* was mainly detected on legs (1.79 times more than other parts of body) (OR: 1.79, CI: 1.17 - 2.71). *Rhipicephalus turanicus* was found on arms 2.82 times (OR: 2.82, CI: 2.00 - 9.94) more than other parts of the body (Table 3). o

Table 1. Demographic characteristics of patients (n=4508) with tick bites

	Gender	Male	Female	Total	%
Living area	Rural	1,713	1,027	2,740	60.78
	Urban	1,112	656	1,768	39.22
Age groups	0-9	375	241	616	13.66
	10-19	666	283	949	21.05
	20-39	760	468	1,228	27.24
	40-64	776	570	1,346	29.86
	≥ 65	248	121	369	8.19
Attachment sites	Abdomen	267	189	456	10.12
	Arms	432	170	602	13.35
	Axilla	250	95	345	7.65
	Back	236	151	387	8.58
	Chest	110	77	187	4.15
	Head and neck	404	373	777	17.24
	Hip	125	120	245	5.44
	Legs	693	358	1,051	23.31
	Perineum	308	150	458	10.16

DISCUSSION

Ticks are obligate blood-sucking ectoparasites of terrestrial vertebrate hosts. Most of the tick species have only limited host specificity; however it is known that there is a variety of factors which influence the host predilection of ticks (25). Although humans are accidental or incidental hosts of ticks (26), tick-bites are frequently seen among farmers, livestock handlers, s^hepherds, slaughterhouse workers, military personnel, picnickers, butchers, veterinarians, and naturalists (6,8,20). The risk of exposure to tick bites is highest especially in edges of forest and in areas with tall grasses and bushes (27). The Tokat province is an endemic region for CCHF (2, 28, 29) and every year over 5,000 tick bite cases have been reported from this province (30,31). By the end

of 2014, 1,908 CCHF cases and 85 deaths (4.45%) were documented in this area. The CCHF cases/deaths were as follows: 2002-2003: 74/5, 2004: 101/4, 2005: 71/4, 2006: 89/4, 2007: 139/7, 2008: 209/6, 2009: 211/10 2010: 162/6, 2011: 258/13, 2012: 168/3, 2013: 209/10, and 2014: 217/13). Bites of *H. marginatum* are common in this region, accounting for 50-70% of all tick bites (28, 32).

In a study contacted by Bursali et al. (30), a total of 5,999 ticks were collected from humans in Tokat province. Out of 5,999 adult ticks collected in 2008, 800 were identified to species level, while the remaining was identified to genus level. Initially, 24 hard ticks infesting humans were found in Tokat province; however, afterwards some specimens detected in their study were re-examined and the

Table 2. Distribution of ticks species related to the age groups of patients

Ticks / age groups	0-9	10-19	20-39	40-64	≥ 65	Total
<i>Argas vespertilionis</i>				1		1
<i>Dermacentor marginatus</i>	14	24	28	28	6	100
<i>Dermacentor niveus</i>		2	4	3		9
<i>Dermacentor</i> nymphs	10	5	6	4	1	26
<i>Hyalomma aegyptium</i>	3	12	17	19	5	56
<i>Hyalomma excavatum</i>	5	16	35	41	10	107
<i>Hyalomma marginatum</i>	324	660	897	1,027	290	3,198
<i>Hyalomma rufipes</i>		1		1		2
<i>Hyalomma</i> larvae	4					4
<i>Hyalomma</i> nymphs	59	38	57	55	21	230
<i>Haemaphysalis erinacei taurica</i>	4		5	2		11
<i>Haemaphysalis parva</i>	89	44	50	25	4	212
<i>Haemaphysalis punctata</i>	12	13	11	17	4	57
<i>Haemaphysalis sulcata</i>	2		2			4
<i>Ixodes frontalis</i>			1			1
<i>Ixodes gibbosus</i>	2	4	1	3		10
<i>Ixodes laguri</i>	1		1			2
<i>Ixodes redikorzevi</i>	3	2	9	12	2	28
<i>Ixodes ricinus</i>	5	6	7	11	6	35
<i>Rhipicephalus annulatus</i>	3		2	3		8
<i>Rhipicephalus bursa</i>	39	60	62	53	14	228
<i>Rhipicephalus sanguineus</i>	2	1				3
<i>Rhipicephalus turanicus</i>	34	61	33	41	6	175
<i>Rhipicephalus</i> nymphs	1					1
Total	616	949	1,228	1,346	369	4,508

total number of ticks infesting humans was corrected as 18 (32). In the present study, a total of 20 tick species infesting humans were identified. It was also found that 62.67% of the patients were males, while 37.33% were females. Similar rates have been found by Sumer (33) (51.8% and 48.2%, respectively); by Erol et al. (20) (54.5% and 45.5%, respectively) and by Over et al. (34) (52.1% and 47.9%, respectively). Previous studies reported that tick bites in children are more common than adolescents, adults and older

person (35,36). In this study, however, we observed that the largest proportion of tick bites were seen in 40-64 age groups (n=1,346, 29.86%), followed by 20-39 years old age group (n=1,228, 27.24%).

Our findings are partially consistent with studies of Erol et al. (20) and Sumer (33). We believe that these findings are most likely associated with the number of residents of Tokat Province in the 40-64 age groups or their intensive agricultural activities.

Table 3. Attachment sites of ticks on the patients

Ticks/Attacment sites	Abdomen	Arms	Axilla	Back	Chest	Head and neck	Hip	Legs	Perineum	Total
<i>Argas vespertilionis</i>						1				1
<i>Dermacentor marginatus</i>	7	22	1	5	2	20	3	35	5	100
<i>Dermacentor niveus</i>	3	1				2	1	2		9
<i>Dermacentor nymphs</i>	2	3	3		2	7	2	3	4	26
<i>Hyalomma aegyptium</i>	4	3	2			2	3	40	2	56
<i>Hyalomma excavatum</i>	8	19	12	8	2	10	2	34	12	107
<i>Hyalomma marginatum</i>	343	377	264	296	136	428	197	804	353	3,198
<i>Hyalomma rufipes</i>				1				1		2
<i>Hyalomma larvae</i>			1			3				4
<i>Hyalomma nymphs</i>	13	31	29	16	20	43	8	27	43	230
<i>Haemaphysalis erinacei taurica</i>	1	4	1			4		1		11
<i>Haemaphysalis parva</i>	14	29	8	10	7	113	3	19	9	212
<i>Haemaphysalis punctata</i>	1	10	1	9	2	23	2	8	1	57
<i>Haemaphysalis sulcata</i>	1	1				1		1		4
<i>Ixodes frontalis</i>								1		1
<i>Ixodes gibbosus</i>		4			1	4			1	10
<i>Ixodes laguri</i>		1						1		2
<i>Ixodes redikorzevi</i>	1	3	3		2	9	1	6	3	28
<i>Ixodes ricinus</i>	4	11		3	4	2	2	7	2	35
<i>Rhipicephalus annulatus</i>		3				4	1			8
<i>Rhipicephalus bursa</i>	34	29	14	26	3	49	18	41	14	228
<i>Rhipicephalus sanguineus</i>						2			1	3
<i>Rhipicephalus turanicus</i>	20	51	6	13	6	49	2	20	8	175
<i>Rhipicephalus nymphs</i>						1				1
Total	456	602	345	387	187	777	245	1,051	458	4,508

Ticks may be found on several anatomical sites of humans but mainly on and around the head and neck area as well as in perineum (37). In the present study it was observed that most of the ticks were found on legs (n=1,051, 23.31%), followed by head and neck (n=777, 17.24%) and arms (n=602, 13.35%). These findings are partially consistent with studies of Gunduz et al (19), Erol et al. (20) and Sumer (33).

There are several studies about the evaluation of tick bites from the different parts of Turkey (19, 20, 33); however, only limited numbers of patients with tick bites were evaluated, e.g., 67 cases in Gunduz et al., (19), 168 cases in Sumer, (33), 161 cases in Erol et al. (20), and 273 cases in Over et al. (34). Only, in a recent study by Kar et al. (38), 1,816 patients were analyzed from the Istanbul province for tick bites. The authors reported that predilection sites of attachment

for *Hyalomma aegyptium* and *D. marginatus* were the extremities and scalp, respectively, while *H. marginatum* and *Ixodes ricinus* were found mainly on the trunk. In the present study, *D. marginatus*, *H. excavatum*, and *H. marginatum* ticks were mainly found on legs, *H. parva* and *R. bursa* on head and neck, and *R. turanicus* on arms. *Argas vespertilionis* and *Ixodes gibbosus* ticks were reported on humans for the first time in Turkey.

CONCLUSION

In the present study, we found that 20 tick taxa (19 species and 1 subspecies) can be infest on humans in the study area. Results of this study also showed that preferred attachment sites of ticks may be changed by demographic properties and age groups of patients. Tick bites are one of the important public

health problems in Turkey, especially in CCHF endemic regions. Accurate and immediate tick removal from the human skin is very crucial to prevention of tick-borne diseases. Despite farmers, livestock handlers, shepherds, slaughterhouse workers, military personnel, picnickers, butchers, veterinarians, and naturalists are at increased risk of tick bites, and subsequently tick-borne disease, anyone who spends any amount of time outdoors in suitable tick habitats, can potentially be exposed to ticks.

For those people it is advised to: (I) wear protective and light colored clothing; (II) use of repellents containing 20-30% DEET (N, N-diethyl-m-toluamide) on the exposed skin areas or 0.5% permethrin on clothing; (III) regularly check clothing and skin and promptly remove attached ticks; and (IV) avoid habitats where ticks are abundant and most active (39,40).

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