Original Article

Biodiversity Indices and Medically Importance of Ticks in North Khorasan Province, Northeast of Iran

Kourosh Arzamani1; *Abedin Saghafipour2; Seyed Ahmad Hashemi1; Hassan Vatandoost3; Mohammad Alavina1,4; Saber Raeghi5; Zakkyeh Telmadarrai6

1Vector-borne Diseases Research Center, North Khorasan University of Medical Sciences, Bojnurd, Iran
2Department of Public Health, Faculty of Health, Qom University of Medical Sciences, Qom, Iran
3Department of Medical Entomology and Vector Control, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran
4Toronto Rehabilitation Centre, University Health Network, Toronto, Canada
5Department of Laboratory Sciences, Maragheh University of Medical Sciences, Maragheh, Iran

*Corresponding author: Dr Abedin Saghafipour, E-mail: abed.saghafi@yahoo.com

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Abstract

Background: Ticks are considered as the main vectors for the transmission of various pathogens such as relapsing fever and CCHF to humans. This study was investigated the biodiversity indices and medically importance of ticks in North Khorasan Province, Northeast of Iran during 2015–2019.

Methods: Specimens were captured from infested ruminants including cows, sheep, and goats. Additionally, tick collections also were performed on non-domesticated creatures such as turtles, rodents, and hedgehogs. Specimens were identified using valid identification keys. Species diversity, species richness and evenness indices have been calculated to estimate species biodiversity of ticks.

Results: A total of 1478 adult ticks were collected. The specimens were from two families: Ixodidae (90.05%) and Argasidae (9.95%), 6 genera and 17 species including: Rhipicephalus sanguineus (55.9%), Rhipicephalus bursa (13.4%), Hyalomma marginatum (9.5%), Hyalomma anatolicum (9.5%), Hyalomma asiaticum (0.2%), Hyalomma aegyptium (0.5%), Hyalomma scapense (1.3%), Hyalomma sp (1.2%), Haemaphysalis sulphata (0.7%), Haemaphysalis erinacea (0.1%), Haemaphysalis inermis (0.1%), Haemaphysalis punctata (0.2%), Haemaphysalis concinna (0.1% Boophilus annulatus (1.2), and Dermacentor marginatus (6.1%) among hard ticks as well as Argas persicus (91.8%) and Argas reflexus (8.2%) amongst soft ticks. Rhipicephalus sanguineus, Rh. Bursa, H. marginatum and H. anatolicum were known as the most frequent species of hard ticks. Tick’s species richness, Shannon diversity index and Simpson index in this area were S= 17, H'= 1.69, D= 0.294 respectively.

Conclusion: Based on tick distribution veterinary authority, public health organizations and other officials should act for implementation of disease prevention.

Keywords: Tick infestations; Ruminant; Geographic information system; Ixodidae; Argasidae

Introduction

There are two major tick (Arthropoda: Arachnida) families: Argasidae and Ixodidae. In these families, some genera and species are able to transmit some important pathogens to humans and livestock. The adult ticks live for some years. In the absence of host, they can survive and may endure hunger for several years. Both male and female ticks are blood sucking and paly as vector of diseases. Some of tick species are able to transmit these agents to the next generation (1). Ticks are blood-sucking ectoparasites of vertebrates, especially wild animals. After a tick bite, people may develop any of flu-like symptoms including fever, weakness, pain and swelling (2). The ticks transmit several bacterial, protozoal, rickaettsial, spirochalal, and viral diseases of humans such as Q fever (Coxiella burnetii), tick-borne relapsing fever...
(Borrelia), rocky mountain spotted fever (rickettsia), Tularemia (pasteurella), Crimean-Congo Hemorrhagic Fever (CCHF) and so on (3). In addition, ticks are able to transmit veterinary pathogens related to babesiosis, theileriosis, Ehrlichiosis and Anaplasmosis in ruminants (4). Although, ticks were not known and familiar to humans for a very long times but their importance in terms of problems in livestock were began from the mid-nineteenth century (5). Currently, there are 896 species of ticks all over the World (6). According to many studies have been performed on fauna of medical important ticks, they are distributed vastly in all regions of the world (7-10). A study on ectoparasites of sheep and goats in the eastern part of the Amhara region, northeast Ethiopia revealed that there are 5 species of ticks including Boophilus decoloratus, Amblyomma variegatum, Amblyomma gemma, Rh. pulchellus, Rh. evertsi on body of sheep and goats. Also, four genera and six species of ticks’ species and treatment of cows, sheep and goats in the Sivas-Zara region, in Turkey were reported (11-12). Previously, many studies on the distribution of tick fauna in Iran have been done (13-19). Asadollahi et al. (2014) have reported the presence of eight tick species in four different genera and they were identified as Hyalomma anatolicum (39%), Rhipicephalus sanguineus (25.4%), Hy. excavatum (14.3%), Hy. asiaticum (6.8%), Haemaphysalis sulcata (3.7%), Hy. scupense (3.4%), Hyalomma sp. (3.4%), Rh. anuulatus (2%) and Hy. dromedarii (2%) in Khuzestan Province, South-west of Iran (20). Ramezani et al. (2014) carried out a study in Meshkinshahr County, Ardabil Province, and reported nine species of ticks including Dermacentor marginatus, D. niveus, Haem. erinacei, Haem. punctata, Hy. anaticum, Hy. asiaticum, Hy. marginatum, Rh. bursa and Rh. sanguineus. Also, Ganjali et al. (2014) reported different species of Ixodidae (hard ticks) from sheep, goats, cattle and camels in Zabol County, the Eastern area of Iran that they were included Hy. dromedarii (17.3%), Hy. schul-zei (1.8%), Hy. marginatum (0.5%), Hy. excavatum (12.60%), Hy. anatolicum (11.2%), Hy. asiaticum (11.0%), Rh. sanguineus (21.2%), Rh. bursa (10.2%) and Rh. turacunis (13.911%) (21-22). The Iranian Center for Disease Control and Prevention (CDC) reported some cases of CCHF infestation in ticks that collected from endemic areas of disease in Iran such as the North Khorasan Province (23). Annually, most cases of CCHF in Sistan and Baluchestan, Isfahan, Fars, Tehran, Khorasan, and Khuzestan are reported in Iran (24). The most recent data indicate a total of 870 confirmed cases of CCHF in Iran, with 126 deaths from 2000 to 2012 (25). North Khorasan Province is one of the transit sites of domestic animals from the eastern borders (Afghanistan and Pakistan) into the Country. Furthermore, ticks are able to transmit different infectious agents to humans and animals, therefore identification of ticks and geographical distribution of them are essential to evaluate epidemiology and risks of human and animal tick-borne diseases. This study was carried out to investigate the biodiversity indices and medically importance of ticks in North Khorasan Province, Northeast of Iran during 2015–2019.

Materials and Methods

Study area
This cross-sectional study was conducted in North Khorasan Province, northeast of Iran, between 36°37'-38°17' N latitudes and 55°53'-58°20' E longitudes with an area of more than 28,434km². The province is bordered by Turkmenistan in the North (Fig. 1). According to the 2011 census in Iran, it includes eight cities with a population of more than 867,000 inhabitants. The province has desert and mountainous areas and receives about 250mm of rainfall annually.

Ticks collection
In this cross-sectional descriptive study, the ticks were collected from infested ruminant in-
cluding cows, sheep, and goats during the period from 2015 to 2019. Additionally, some specimens collected from turtle, rodent, and hedgehog. The tick samples were collected from 53 different areas in the province. Specimens were performed by examining the whole body including, ears, abdomen, tail, perianal regions, milk gland area, and back of the animals. Collected ticks were placed in separate dry and labeled vials and transferred to the laboratory of Vector-borne Diseases Research Center, North Khorasan University of Medical Sciences.

**Determination of the Species Diversity**

All specimens were identified based on morphological characteristics and the keys given by Estrada-Pena (2004) in the laboratory of medical entomology, Tehran University of Medical Sciences (26). All statistical analyses were done by excel software Ver. 2010. and Alpha diversity indices were calculated using an online biodiversity calculator (27).

**Results**

In this study, 515 (55.2%), out of the total 933 inspected sheep, goat and cow were infested with ticks. A total of 1478 ticks (770 males and 708 females) were collected (Table 1). The identified tick specimens belonged to two families: Ixodidae (90.05%) and Argasidae (9.95%), 6 genera and 17 species including concerning Ixodidae families, *Rh. sanguineus* (55.9%), *Rh. bursa* (13.4%), *Hy. marginatum* (9.5%), *Hy. anatolicum* (9.5%), *Hy. asiaticum* (0.2%), *Hy. aegyptium* (0.5%), *Hy. scupense* (1.3%), *Hy. sp* (1.2%), *Haem. sulcata* (0.7%), *Haem. erinacea* (0.1%), *Haem. inermis* (0.1%), *Haem. punctata* (0.2%), *Haem. concinna* (0.1), *B. annulatus* (1.2), and *D. marginatus* (6.1%) (Table 1). *Rhipicephalus* (69.3%) followed by *Hyalomma* (22.2%) were the predominant tick genus of hard ticks. With regards to the seasonal variation, the highest and lowest tick infestation was recognized in spring and winter, respectively.

The results show that out of soft ticks only *Ar. persicus* (91.8) and *Ar. reflexus* (8.2) were collected in this region (Table 2). All species of hard ticks were collected from sheep except *Hy. Aegyptium* and *Haem. concinna*. Collected species based on the hosts are shown in Table 3.

Some of the most important Alpha diversity indices including, Species richness, Shannon-Wiener index, Simpson index, Shannon Evenness index (Equitability index), Buzas and Gibson's evenness index, Berger-Parker dominance index, Menhinick richness index, and Margalef richness index were calculated (Table 4).

**Table 1.** The frequency of hard ticks by gender in North Khorasan Province, Northeast of Iran during 2015–2019

| Species        | Male | Female | Total | %   |
|----------------|------|--------|-------|-----|
| *Rh. sanguineus* | 404  | 340    | 744   | 55.9|
| *Rh. Bursa*    | 87   | 91     | 178   | 13.4|
| *Hy. marginatum* | 80  | 47     | 127   | 9.5 |
| *Hy. anatolicum* | 70  | 56     | 126   | 9.5 |
| *D. marginatus* | 41   | 40     | 81    | 6.1 |
| *Hy. scupense* | 8    | 10     | 18    | 1.3 |
| *B. annulatus* | 0    | 16     | 16    | 1.2 |
| *Hy. Sp*       | 0    | 16     | 16    | 1.2 |
| *Haem. sulcata* | 4    | 6      | 10    | 0.7 |
| *Hy. Aegyptium* | 3   | 3      | 6     | 0.5 |
| *Hy. asiaticum* | 3   | 0      | 3     | 0.2 |
| *Haem. punctata* | 2  | 1      | 3     | 0.2 |
| *Haem. erinacea* | 0  | 1      | 1     | 0.1 |
| *Haem. inermis* | 0   | 1      | 1     | 0.1 |
| *Haem. concinna* | 1  | 0      | 1     | 0.1 |
Table 2. The prevalence of soft ticks in North Khorasan Province, Northeast of Iran during 2015–2019

| Host       | Visit | Rh. sanguineus | Rh. Bursa | Hy. marginatum | Hy. anatolicus | D. margaritus | Hy. scapense | B. annulatus | Hy. sp | Haem. sulcata | Hy. aegyptium | Hy. asiaticum | Haem. punctata | Haem. erinacea | Haem. inermis | Haem. concinna | Total of all specimens |
|------------|-------|----------------|-----------|----------------|----------------|---------------|--------------|--------------|--------|--------------|---------------|---------------|----------------|----------------|----------------|---------------|----------------|---------------------|
| Sheep      | 812   | 473            | 107       | 107            | 91             | 81            | 12           | 16           | 10     | 8            | 0             | 3             | 3             | 1             | 1             | 0                   | 913                      |
| Goat       | 81    | 267            | 67        | 2              | 2              | 0             | 0            | 0            | 2      | 0            | 0             | 0             | 0             | 0             | 0             | 0                   | 340                      |
| Cow        | 40    | 0              | 2         | 18             | 33             | 0             | 0            | 0            | 0      | 0            | 0             | 0             | 0             | 0             | 0             | 0                   | 59                       |
| Turtle     | 2     | 0              | 0         | 0              | 0              | 0             | 0            | 0            | 6      | 0            | 0             | 0             | 0             | 0             | 0             | 0                   | 6                        |
| Rumbomis meriones | 1 | 0          | 0         | 0              | 0              | 0              | 0            | 0            | 4      | 0            | 0             | 0             | 0             | 0             | 0             | 0                   | 4                        |
| Hedgehog   | 5     | 4              | 2         | 0              | 0              | 0             | 0            | 0            | 0      | 0            | 0             | 0             | 0             | 0             | 0             | 1                   | 7                        |

Total specimens: 703

Table 3. Species and numbers of hard ticks on different host collected in North Khorasan Province, Northeast of Iran from 2015 to 2019

| Species       | Male | Female | Total | %   |
|---------------|------|--------|-------|-----|
| Ar. persicus  | 59   | 76     | 135   | 91.8|
| Ar. reflexus  | 8    | 4      | 12    | 8.2 |

Total of all specimens: 147

Table 4. Biodiversity and species richness indices of hard ticks in North Khorasan Province, North east of Iran from 2015 to 2019

| Index                              | Computation                                                                 | Amount |
|------------------------------------|----------------------------------------------------------------------------|--------|
| specimens                          | Total number of specimens                                                 | 1478   |
| Species richness (S)               | The number of species                                                     | 17     |
| Shannon-Wiener index (H’ )         | $H' = \sum_{i=1}^{S} p_i \ln(p_i)$                                      | 1.69   |
| Simpson Index                      | $\frac{\sum_{i=1}^{S} n_i(n_i - 1)}{N(N - 1)}$                           | 0.294  |
| Shannon Evenness Index (Equitability Index) | $\frac{H}{\ln(S)}$                                    | 0.595  |
| Buzas and Gibson's Evenness Index  | $\frac{e^{-\sum_{i=1}^{S} p_i \ln(p_i)}}{n_{\text{max}}}$               | 0.317  |
| Berger-Parker Dominance Index      | $\frac{N}{S}$                                                             | 0.503  |
| Menhinick Richness Index           | $\frac{\sqrt{\sum_{i=1}^{S} n_i}}{S}$                                  | 0.442  |
Discussion

Understanding the fauna species and geographical distribution of ticks will provide appropriate strategies to control and prevention of the tick-borne diseases (28). North Khorasan Province is bordered with Turkmenistan. This province is a suitable area for agricultural production and animal husbandry (29). A considerable large number of sheep, goats, cows and so on graze on pastures of the province and therefore are at a greater risk of tick infestation and their borne diseases (30). In this study, most of ticks were collected on the body of sheep, goats and cows, but some of other animals such as turtles, some rodents of Rhombomys and Meriones spp. and Hedgehog were infested with ticks. Most of ticks such as Ixodes, Dermacentor, Rhipicephalus, Haemaphysalis have at least three hosts. Telmadarrai et al. have showed that Rh. sanguineus, Hy. marginatum, Hy. anatolicum, Hy. asiaticum and Hy. dromedarii were known as the most frequent species which were positive for CCHF virus in endemic areas of Iran (23). Most of these proven vectors of CCHF in Iran were collected and identified in North Khorasan in this study. In this present study, 17 species of ticks were identified including Rh. sanguineus, Rh. bursa, Hy. marginatum, Hy. anatolicum, Hy. asiaticum, Hy. aegyptium, Hy. scupense, hyalomma sp, Boophilus annulatus, Dermacentor marginatus, Argas persicus, Ar. reflexus. It has been
shown that the tick infestation on cattle included *Haem. parva* (33.8%), *Dermacentor marginatus* (2.8%), *Boophilus annulatus* (21.1%), *Haem. concinna* (15.5%), *Hy. marginatum* (19.7%) and *Rh. bursa* (7%). Besides that, many studies on tick fauna have been conducted in different areas of Iran. For instance, in West Azarbaijan, 7 genera and 15 species were identified including *Rhipicephalus, Hyalomma* and *Orithodoros* and *O. lahorensis, H. marginatum, H. asiaticum, Rh. bursa* had the highest frequency (31). Nabian et al. reported the presence of fourteen tick species were identified as *Hy. anatolicum* (5.23%), *Hy. marginatum* (20.34%), *H. detritum* (3.48%), *Haemaphysalis punctate* (12.79%), *Haem. Parva* (0.58%), *Haem. concinna* (0.58%), *Haem. choldokovsky* (6.97%), *Ixodes ricinus* (2.32%), *Rh. sanguineus* (19.76%), *Rh. bursa* (4.65%), *Boophilus annulatus* (9.88 %), *Dermacentor niveus* (6.39%), *D. marginatus* (1.74 %) and *O. lahorensis* (5.23%) in Ardabil, north west of Iran (18). In this research, the genera *Rhipicephalus* (69.3%) and *Hyalomma* (22.2%) were the most predominant ticks. Out of them *Rh. sanguineus, Rh. bursa*, *Hy. marginatum* and *Hy. anatolicum* had highest frequencies among hard ticks. A similar study was also reported from Meshkinshahr (a northwest area in Iran) in 2009 by Hosseini vasoukolaei et al. in which most of the ticks belonged to *Rhipicephalus* and *Hyalomma* genus (19). Based on the results of the Mazlum (1971) carry out on ticks *Rh. bursa, Rh. sanguineus, Hy. anatolicum* were the most common ticks in Iran that are widespread in various areas of the country (15). Nabian et al. showed that *Rh. sanguineus* was the dominant tick species on body’s seep in Mazandaran Province, North of Iran (18). *Rhipicephalus bursa* and *Rh. sanguineus* are very important in terms of health and medicine in Iran because they have basic role in the transmission of pathogens in sheep. The pathogens that are transmitted by these tick species are *Babesia canis, Ba. equi, Ba. caballi, Theileria ovis, Anaplasma marginale* and *Rickettsia oina* (32). The *Hyalomma* ticks are most common ticks in Iran has been isolated from different domestic animals. Mazloum studies indicated that *Hy. anatolicum* is one of the prevalent ticks in Iran. *Hyalomma anatolicum* can transmit many infectious agents of humans such as *Theileria anatolica*, *Theileria parva, Ba. caballi, Q fever* and CCHF. Previous epidemiological studies in Khorasan Provinces have demonstrated that CCHF disease is one of endemic disease in this region (23, 25). It also has an important role in transmission of *Theileria Herci* to sheep in Iran with regards to the seasonal variation, the highest and lowest tick infestation was recognized in spring and winter, respectively (33). Other studies have been indicated the highest tick infestation distribution was spring and summer (16, 34-35). In principle, ticks are most active in hot seasons and humidity. In these areas as due to livestock grazing season (spring and summer when the heat is suitable for complete life cycle of ticks) and putting them in the pasture, the tick distribution will be mainly influenced by rainfall (36). Six *Haemaphysalis* species have been recorded by different authors in Iran and three species of them have been reported previously from North Khorasan Province (37). In the current study we report five species of this genus, so all species of this genus which reported from Iran collected from North Khorasan Province. There are only limited studies in Iran in which the authors calculated the biodiversity indices of ticks. Furthermore, to the best of our knowledge, there has not been any report on biodiversity of ticks in North Khorasan Province and this is the first work that provides a numerical index on biodiversity of ticks in this region. The result showed tick’s species richness, Shannon diversity index and Simpson index in this area were higher than some other regions in the country (38-41).

**Conclusion**
The present study showed a high degree of diversity of ticks in North Khorasan Province, northeast of Iran. Some of the most medically and veterinary important species of ticks were collected from this region. It is important to awareness of fauna and the distribution of ticks in order to apply effective tick control programs and prevention of the disease in the region. We declare that we have no conflict of interest.

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