Original Article

Diversity of Hard Tick Populations and Their Geographical Variations in Northwestern Iran

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Abstract

Background: Ticks are forced vertebrate ectoparasites, including humans, and are vectors of serious diseases such as Crimean Congo Hemorrhagic Fever, Relapsing Fever, and various forms of encephalitis. Spatial assessment of the prevalence of ticks and detection of high risk areas for tick-borne disease transmission and evaluation of ecological measures are key aims of this research.

Methods: Ticks were collected using standard methods from 27 villages in the region of Sarab County in north-eastern Iran during the four seasons of 2018–2019 and identified using valid keys. The calculations of indices for biodiversity were based on the Margalef index, Shannon-Weiner index and Simpson index. R2.15 Statistical software was used for statistical analysis of indices of biodiversity, and ArcMap10.4.1 software, IDW and GeneralG methods were used. Analysis were used to investigate spatial distribution and to determine important tick hotspots.

Results: A total of 2500 animals surveyed, 35% of them were infected. In total, 1416 ticks were caught, 74.6 %, 23.9% and 1.4 % were adult, nymph and larvae respectively. 94% of the ticks were hard ticks including 6 genera. According to the Margalef diversity index, the highest species biodiversity was related to summer (1.4234), and the lowest was related to winter (0.7379),

Conclusion: Large hotspot area was found in the central part of the study area. The area of study was very prone to tick-borne disease transmission in terms of tick diversity and tick species richness. Tick-borne disease control is an important measure.

Keywords: Ticks; Species biodiversity; GIS; Iran

Introduction

Ticks are the most important medical and veterinary ectoparasites and are one of the most important arthropod groups (1). Ticks belong to the order of metastigmata which feeds on animals and humans and play an important role in the transmission of the disease. This order is composed of two common Argasidae and Ixodidae families (2). There are about 899 hard tick species, and 185 soft tick species (3). In Iran 14, 5, 8, 11, 1 and 3 hard tick species of *Hyalomma*, *Rhipicephalus*, *Ixodes*, *Haemaphysalis*, *Bophilus* and *Dermacentor* has been recorded respectively. 10% of ticks feed on domestic animals, particularly cattle, sheep and goats. These are of major health significance due to the conditions that cause human and animal diseases (4-6). Tick-borne diseases are among the most common emerging and re-emerging diseases in recent years that have spread to different geographic areas (7). Several diseases including tick-borne fever, Rocky mountain spotted fever, Q fever, Lyme disease, and Crimean-Congo Hemorrhagic Fever (CCHF) are among the known tick-borne human diseases

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(8-10). Regardless of the importance of ticks in spreading different pathogens, its population structure, relative abundance and frequency, hosts type, habitats, ecology and diversity of species must be regulated. The study of tick biodiversity using various scientific indices seems to be very helpful (11-12). The aims of this study was to determine the status of species diversity and richness of hard ticks and their geographical distribution in the livestock of Sarab region in northwestern Iran using Simpson, Shannon Weiner, Margalef Indices and GIS software indicators.

Materials and Methods

Study area

The county of Saraba is located in the province of East Azerbaijan, Iran. The capital of the county is Sarab. The county population at the 2006 census was 132,094, out of 31,977 families. The county is subdivided into two districts: the Central District and Mehraban District. The county has four cities: Sarab, Mehraban, Sharabian, and Duzduzan. At the 2006 census, it had a population of 42,057 in 11,045 families. Livestock and animal husbandry has traditionally been common in most rural households (Fig. 1).

Tick collection

A total of 27 villages were selected from 9 districts. Ten locations were selected in each village (human, animal and livestock storage areas, traditional and modern grain deposits, farmland attached to the village and preferably attached to active livestock sables, estimated at 300 places). During the study, 441000 cattle (331000 sheep and goats) and (95000 cows and oxen) (8000 horses) and (7000 donkeys) were in the Sarab County. Therefore, ticks from a total of 2500 livestock were collected in this study, of which 75% of the sample size were sheep and goats, 23% were cows and oxen, and 2% were horses and donkeys. Valid keys were used to identify species of the ticks (13). This experiment was carried out under the guidance of the Ethics Committee of the Tehran University of Medical Sciences (IR.TUMS.SPH.REC. 1398.058).

Biodiversity Indices

Different indices of biodiversity were used in this study including species richness (Margalef's richness index), species diversity (Simpson's and Shannon Weiner's index).

Calculation of tick dispersion

Arcmap 10.4.1 software and interpolation with Inverse Distance Weighted (IDW) were used to analyze tick dispersion and to assess the distribution of disease vectors hot spots. The General G formula was used to determine the pattern of distribution of all CCHF disease species and main vectors including *Hyalomma marginatum* and *H.anatolicum* in the study region.

$$G = \frac{\sum_i^N = 1 \sum_j^N = 1 Wij\ xi\ xj}{\sum_i^N = 1 \sum_j^N = 1 xi\ xj}, j \neq i$$

Results

For a total of 2500 animals examined, ticks infested 763 animals (30.5 %). In total, 300 stalls and stables were inspected in 27 villages, 75 of which were infested with ticks (25%). A total of 1,416 ticks have been collected, of which 30% were male, 44.6% were female, 23% were nymphs and 1.4% were also tick larvae. Of the 94% of the ticks collected, 11 species have been classified as hard ticks, belonging to 4 genera: *Hyalomma* (65%), *Haemaphysalis* (11%), *Rhipicephalus* (6%), and *Dermacentor* (12%). Three species of soft ticks from two genera of *Ornithodoros* and *Argas* were identified (Table1).

Sheep and goats with 58.68%, cows and ox with 13.23%, horses and donkeys with 2.14% respectively were the most infested animals with ticks. In terms of age, ticks were observed more in animals under 3 years of age (49 %) and the lowest infection (10 %) was observed in ani-

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mals over 7 years of age, and the most infested section of the animals were sub-tail (29.89 %) and animal ears (25.69 %).

Biodiversity and species richness

Tick diversity and species richness were calculated on the basis of two factors: seasonal activity and the host. The Margalf species richness index showed that the highest richness was related to summer (1.4234), and the lowest richness was related to winter (0.7379). The species diversity of ticks collected at different seasons in the research region were also significantly different ($P< 0.05$). The highest species diversity was related to summer (2.1709), and the lowest species diversity was related to winter (0.781), according to the Shannon-Wiener index measurements. The index of evenness of ticks collected during different seasons in the research region was significantly different ($P< 0.05$). The evenness index analysis found that the highest uniformity was in relation to the summer season (0.9428) and the lowest uniformity was in relation to the autumn season (0.4428). The Simpson species diversity index also showed that the highest diversity was correlated with summer (0.8622), and the lowest diversity was associated with autumn (0.3549). The results of this study found that the species richness of ticks collected in the Sarab Region during the first and second six months of the year had a significant difference ($P< 0.05$), and the first six months of the year had higher and more diversity and richness of species than the second six months of the year (Table 2).

According to the Simpson Species Diversity and the Shannon-Wiener Indices, the findings of species diversity and richness based on different hosts showed that the highest species diversity among the studied hosts were correlated to sheep and goats (0.881and 2.333), and the lowest species diversity was found in horses and donkeys (0.715 and 1.399) respectively. There was a significant difference (Table 3) in the evenness index for livestock hosts that ranging from the highest for sheep and goats (0.9096) to the lowest for cows and oxen (0.7945).

Spatial distribution

The most ticks were collected in the villages of Dozduzan (6.43%), Sharabian (6.28%), Mehraban (6.12%), Kelian (5.6%), Ardha (5.54 %) and Mahin Bijand (5.3%) and the lowest in Dichan village (2.45%). In Mehraban District the highest spatial dispersion as well as the highest density of hard and soft tick species was found.

The findings of the interpolation study (IDW) revealed that there was a large hotspot area in the central part and a small hotspot in the northwestern part of the county of Sarab in terms of tick abundance, both of which were high-risk areas in the Mehraban Region with 4596 and 1150 households in population (Fig. 2).

The results of the analysis of spatial distribution patterns for all species and for two species of *H. marginatum* and *H. anatolicum* using the General G index showed that all species were distributed randomly in all areas of study and did not have a specific pattern such as cluster or regular (Fig. 3 and Table 4).

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Table 1. The total number of ticks collected from hosts and residential places in Sarab County, 2018–2019

Family	Genus	Species	Hosts				Adult ticks				
			Cow and Ox	Sheep and Goat	Donkey and Horse	Shelters	Male	Female	Nymph	Larva	Total
Ixodidae	*Hyalomma*	*H.anatolicum*	11	17	0	0	13	33	0	0	46
	Hyalomma	*H.marginatum*	52	69	12	0	206	111	2	0	319
	Hyalomma	*H.scupense*	12	22	0	0	35	25	0	0	60
	Hyalomma	*H.sp*	39	67	2	0	2	141	0	0	143
	Hyalomma	*H.asiaticum*	8	18	2	0	38	17	0	0	55
	Hyalomma	*larva*	0	2	0	0	0	0	0	18	18
	Hyalomma	nymph	32	70	7	0	0	0	279	0	279
	Haemaphysalis	*He.sulcata*	4	65	0	0	17	87	0	0	104
	Haemaphysalis	*He.numidiana* (*erinacei*)	4	14	0	0	13	18	0	0	31
	Haemaphysalis	*He.concinna*	0	19	0	0	0	23	0	0	23
	Rhipicephalus	*R.sanguineus*	8	21	4	3	22	54	0	0	76
	Rhipicephalus	*R.bursa*	8	6	0	0	10	10	0	0	20
	Dermasentor	*marginatus*	42	76	2	0	64	105	0	0	169
Argasidae	*Ornithodoros*	*O.lahorensis*	0	42	0	63	21	65	434	0	520
	Argas	*A.reflexus*	4	2	0	2	4	3	0	0	7
	Argas	*A.persicus*	0	0	0	7	0	13	29	0	42
			224	510	29	75	445	705	744	18	1912

Table 2. Biodiversity and species richness of ticks captured in the studied areas of Sarab County, based on seasonal activity, 2018–2019

	Spring	Summer	Fall	Winter
Shannon-Wiener index	2.1267	2.1709	0.7934	0.781
Simpson's index(D)	0.1449	0.1378	0.6451	0.6385
Simpson's index(D-1)	0.8551	0.8622	0.3549	0.3615
Evenness Index	0.9236161	0.942812	0.4428048	0.485265
Margalef diversity index	1.3702635	1.423474	0.8287641	0.7379352

Table 3. Diversity and species richness of ticks caught separately by the host in the study areas in Sarab County, 2018–2019

	Cow and Oxen	Goat and sheep	Donkey and Horse	Stables
Shannon-Wiener index	1.905	2.333	1.399	0.447
Simpson's index(D)	0.198	0.119	0.285	0.787
Simpson's index(D-1)	0.802	0.881	0.715	0.213
Evennes Index	0.7945	0.9096	0.8693	0.3224
Margalef diversity index	1.807	1.708	1.077	0.483

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Table 4. The results of hot-spot and autocorrelation analysis of tick species in Sarab County, East Azerbaijan, north-west of Iran, 2018–2019

	General G Summary		Global Moran's I Summary		
Species	**Observed General G**	**Expected General G**	**Variance**	**z-score**	**P**
All species	0.000015	0.000016	0.000000	-0.357036	0.721065
Hyalomma marginatum	0.000014	0.000016	0.000000	-1.464457	0.143069
Hyalomma antalicum	0.000013	0.000016	0.000000	-0.911540	0.362011

Fig. 1. Map of studay area, Sarab County, East Azerbaijan Province, Iran

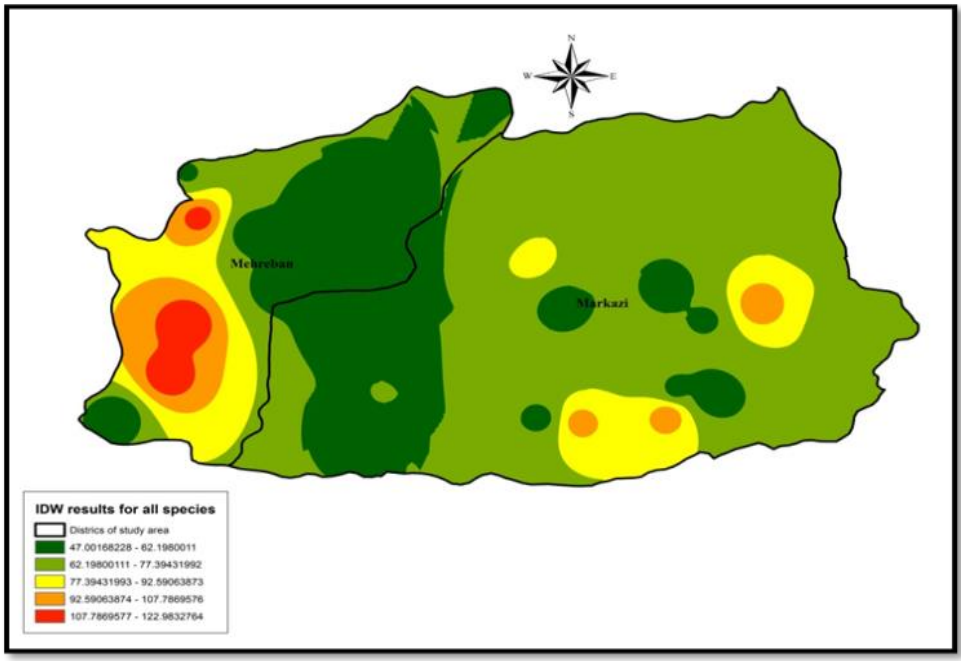

Fig. 2. General distribution of ticks caught in the study areas of Sarab County, 2018–2019

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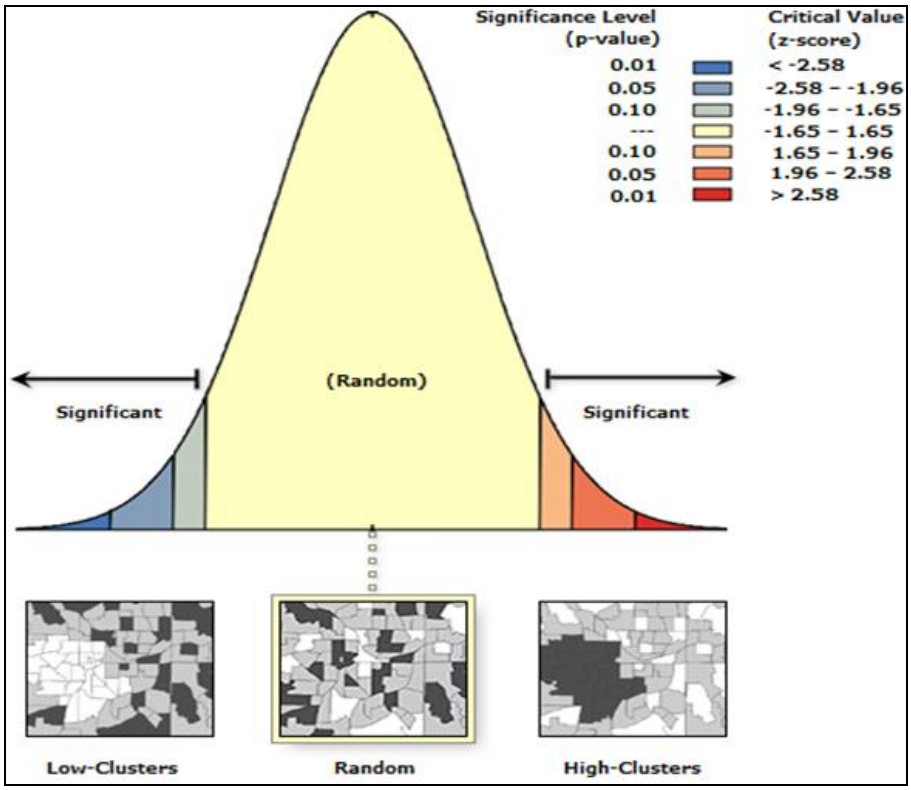

Fig. 3. Results of scattering pattern analysis of ticks caught in Sarab County, East Azerbaijan, northwest of Iran, 2018–2019

Discussion

In this study, 2500 different livestock were studied and 30.5% of livestock and 25% of stalls and stables were infested with different ticks. This is one of the first studies to include more samples and tick populations in Iran. In related surveys, the infestation rate of ticks was 11%, 9.37%, 43% and 24% in northwest, west and north Iran (14-16). The highest frequency of captured ticks was related to spring (37.24%) and the lowest frequency was related to winter (11.83%) and this indicates the seasonal behavior of ticks in the mountainous and northwestern regions of Iran, mainly in spring, which can be attributed to the Nasiri study in Abdanan County, Ilam Province (17). Nonetheless, more seasonal tick activity was recorded in summer and autumn in areas such as Golestan Province northern Iran, which have ecologically different climates and have lower altitudes and higher temperatures than the mountainous areas (18). However, more seasonal tick activity was recorded in summer and autumn in areas such as Golestan Province northern Iran, which have ecologically different climates and have lower altitudes and higher temperatures than the mountainous areas (18). In the study area, the tick biodiversity, according to the Shannon-Wiener index, was found to be moderate to high (2.432) and the biodiversity index of Simpson (D-1) was also determined to be 0.856. The dominant species in the area therefore had a great diversity. We found the highest richness of ticks captured from cows and oxen and the lowest from horses and donkeys, according to the Margalef species richness index. The findings of this research did not confirm previous studies in the province of Golestan (18) that found more sheep species

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compared to cows. The diversity of the species was highest in autumn, and lowest in winter. The findings of this research were consistent with study for seasonal ticks activities in northwestern Iran (19).

Six genera and 14 species of hard and soft ticks were collected in this research, of which the dominant genus was *Hyalomma* (48.12%). The findings of this research revealed that the region was prone to diseases like CCHF. According to the study in 2004 (unpublished data), in the eastern province of Azarbaijan, northwestern Iran, the *Hyalomma* genus was confirmed to have the highest abundance (52.81%) which was consistent with recent study (unpublished data). Interestingly, in a survey conducted in West Azerbaijan Province, 7 tick species with more abundant in the *Hyalomma* genus (unpublished data) were reported to have the highest abundance and distribution, as our recent research did. Based on the findings of this study and the above research, it can be inferred that the provinces located in northwestern Iran including East Azerbaijan, West Azerbaijan and Ardabil had a high abundance of *Hyalomma* species and were ecologically suitable habitat for this tick.

Identifying high-risk and low-risk disease and vector areas allows to make appropriate decisions on the management of the vectors and diseases transmitted by them, and to arrange for the control and preventive management of the region. Throughout this research, the spreading of ticks as the vectors was found randomly around the region, but internal analysis revealed that the high-risk area of ticks was very high in the west of the study area as well as being a large hotspot. The high population of livestock and livestock occupation which have been the key hosts of ticks were some of the factors involved in this region. Therefore, in terms of tick-borne diseases, this area is indeed one of the high-risk areas and control measures should be taken to prevent bites of ticks and diseases that they transmit.

Conclusion

The main method of application of pesticides against ticks are dipping method, pour-on and sometime oral administration by systemic acaricides in Iran as well as in Sarab County, Eastern Azerbaijan Province. The findings of the recent study indicate that the number of tick species caught in this region was varied due to the large abundance of livestock in Sarab County and the wealth of animal and livestock husbandry practices and the production of dairy products. Because of the small scale of the county it reveals the richness of different species of ticks in this region. In the study of tick diversity in the research region among different hosts, it was observed that all the indices observed on sheep and goats have a comparatively higher value that the ticks were more associated with long hair hosts, so periodically picking the wool and hair of these hosts may be a way to reduce tick activity. Throughout this research, the spreading of ticks as the vectors was found randomly around the region, but internal analysis revealed that the high-risk area of ticks was very high in the west of the study area as well as being a large hotspot.

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