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Defining and comparison of biodiversity components of hard ticks on domestic hosts at Highland, Woodland and Plain in Northern Iran

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Abstract. The aim of the present study was to compare the biodiversity patterns of tick species in the Northern Iran between June 2007 and June 2008. 41 villages within three locations, viz. highland, woodland and plain, were selected using cluster randomized sampling method. A total of 504 sheep and 162 cattle visited monthly for a period of 12 months for the presence of ticks. Estimated richness was calculated using rarefaction curves and richness estimators. Defining the contribution of biodiversity components was adjusted using SHE analysis. A total of 1,231 hard ticks (consisting of 6 genera and 6 species) were collected. *Rhipicephalus bursa* (in highland and plain) and *Boophilus annulatus* (in woodland) were identified as eudominant species with the most abundance. Seasonal activity of the ticks varied among species. Rarefaction curves showed that summer and spring had the highest species diversity and richness in different locations. SHE analysis revealed that evenness had the greatest effect on the changes of species diversity and also showed that all regions are mainly occupied by common species. The study demonstrated that the seasonal fluctuations of *R. bursa* have a significant impact on the changes of species diversity.

INTRODUCTION

Hard ticks are important obligatory ectoparasites of humans and livestock. They are serious vectors of various pathogens and infectious diseases to humans and animals (Bursali *et al.*, 2010). Considering the public health and economic impacts of hard ticks, control strategies should be much better defined (Jongejan and Uilenberg, 2004; Bursali *et al.*, 2010). Control measures of hard ticks rely greatly on the knowledge of identification, ecology, distribution and seasonal activity of hard ticks (Žákovská *et al.*, 2013; Torina *et al.*, 2014). These factors can be defined by ecologists in terms of species diversity (Agrawal and Gopal, 2013; Flombaum *et al.*, 2014). Vectors diversity and its changes can potentially impact on the exposure risk of human to tick-borne diseases (Molyneux, 2008; Stearns, 2009), therefore study on the vectors bio-diversity is very important; especially in the scale of time and space. At a glance, species richness (*S*), evenness (*E*) and heterogeneity (Shannon index or *H'*) indices are different aspects of species diversity in any community (Allan *et al.*, 2013). Species richness is a fundamental measurement of diversity but it is highly sensitive to different sampling sizes and as a result, comparisons of species richness may produce inaccurate decisions

(Gotelli and Colwell, 2001). In such cases, an efficient method for solving this problem is to standardize the sample sizes using rarefaction curves (Melo *et al.*, 2003). Rarefaction curves can interpolate different richness to smaller sample sizes and compares observed richness (Chao *et al.*, 2014). Evenness is defined as how evenly the individuals in the community are distributed (Heip *et al.*, 1998) and strongly and positively correlated with species richness and Shannon index (Lamb *et al.*, 2011). For a better interpretation of the relationships between species richness, Shannon index and evenness, SHE analysis is a good solution. This graphical method identifies the fluctuations of these components and distinguishes the patterns of species distribution including: log-normal, log-series or broken stick model (Magurran, 2004; Wilson *et al.*, 2012). Therefore, using rarefaction curves and SHE analysis provide a better understanding of the process changes of biodiversity in different time or spatial scales.

In Iran, occurrence of ixodid ticks has been reported by some researchers (Yakhchali *et al.*, 2011; Ramezani *et al.*, 2014; Ebrahimzadeh *et al.*, 2015), but the comparison of the biodiversity parameters is rarely performed. Mazandaran province, northern Iran has a diverse seasonal climate and rich vegetation and consequently the province is prone for the livestock industry, so offer ideal opportunities to study the ticks' diversity. Therefore, the main purpose of this study is how many species are indeed present in the study site using rarefaction curves. Another goal is to analyze our data set by SHE technique to determine the community structure of hard ticks. In addition, richness estimators including first and second order Jackknife, Chao1 and Chao2 were calculated using data sets to estimate the species richness (Gotelli *et al.*, 2013). Finally, the comparison of diversity components was performed for a better understanding of the effects of different geographical areas, hosts and seasons on the ticks' diversity.

MATERIALS AND METHODS

Study area

Sari County (3685 km^2; 36°34'4"N, 53° 3'31"E), the capital city of the Mazandaran province, Northern Iran, is located to the south of the Caspian Sea and north of the Alborz mountain range, at an altitude between 10 m to 3000 meters above sea level. The climate is mild to temperate and hot (average temperature 16.7) with rainfall (average 690 mm) mostly in the winter, and a little in summer (http://en.climate-data.org/location/ 764529/). The county has three towns, five districts, 15 rural districts and 470 villages, and comprised of three distinct geographical areas, i.e. highland, woodland and plain. The climatic conditions of the plain and woodland are hot and humid in summer and mild and wet in winter while in highland the climate is mild with short summer and cold winter. Plain is situated between altitudes of 10-450 meters above sea level with vegetation including different crops and various shrubs. Woodland vegetation mainly includes deciduous trees at an altitude of 1800 to 2500 meters, while highland is situated between altitudes 1500-3000 meters above sea level, away from the Caspian Sea and its vegetation including grass, shrubs and herbaceous plants. The rural population in the highland and woodland practice mainly animal husbandry (sheep and cattle), while in the plain areas agriculture (rice and citrus) is the main occupation followed by animal husbandry.

Data and tick collection

Cluster randomized sampling was used for specimens collection. Based on the natural geography of the study area, three clusters of plain, woodland and highland areas were selected. Within the clusters, random sampling method was applied to select 15, 14 and 12 villages in highland, plain and woodland areas, respectively. According to the provincial veterinary organization, there are 99,070 cattle and 388,685 sheep in Sari County. The minimum sample size required was calculated according to systematic sampling method (Thrusfield, 2007). A total of 504 sheep and 162 cattle were randomly selected from 17 sampling sites (herds) in the highland (cattle=54; sheep=280), woodland (cattle=66; sheep=76) and plain (cattle=42; sheep=148) during June 2007 to June 2008. Tick specimens were collected from the whole body of each animal using time-series methods, i.e. once a month (Murray, 2003). The specimens were placed in separate appropriately labeled tubes and transferred to the Entomology laboratory, Faculty of Health, Mazandaran University of Medical Sciences and identified using appropriate keys (Hoskins, 1991, Keirans and Litwak, 1989, Walker, 2003).

Categories of Dominance

To evaluate the dominance structure of ticks, Heydemann's classification was used. This classification has five levels of dominance: eudominant species – those making up more than 30% of all the specimens caught, dominant – 10–30%, subdominant – 5–10%, rare – 1–5% and subrare – less than 1% (Heydemann, 1955).

Statistical analysis

Biodiversity indices

Two of the best known species richness indices including Margalef's $(D_{Mg}=\frac{S-1}{lnN})$ and Menhinick's ($D_{Mn}=\frac{S}{\sqrt{N}}$) indices were calculated. For assessing the variance of tick species-abundance distribution Simpson's dominance (D), Simpson's Diversity (1-D) and Berger-Parker dominance indices were used (Table 1). For characterizing tick diversity in a community Shannon's H' and Shannon's equitability (Pielou's index of evenness; E_H) indices were calculated (Table 1). Diversity t test was performed by comparison of the Shannon and Simpson diversities in two samples (Jacobson *et al.*, 2011).

Table 1. List of richness corrected estimators, dominance and heterogeneity indices

Richness corrected estimators	Formula	Descriptions	Reference
Chao 1 estimator	$S_{Chao1} = S + \frac{F_1^2}{2F_2}$	It is based on the number of rare species in a sample	(Chao, 1984)
Chao 2 estimator	$S_{Chao2} = S + \frac{Q_1^2}{2Q_2}$	It is based on the number of species found in just one sample and the number of species found in exactly two	(Chao, 1987)
Jackknife estimators 1	$S_{jackknife1}= S_{obs} + \frac{Q_1(m-1)}{m}$	depends only on the unique species (species found in only one sample)	(Gotelli *et al.*, 2013, Gotelli and Colwell, 2011)
Jackknife estimators 2	$S_{jackknife2} = S_{obs} + \frac{Q_1(2m-3)}{m} - \frac{Q_2\ (m-2)^2}{m\ (m-1)}$	depends only on the unique and the duplicates species (species found in exactly two samples)	(Gotelli *et al.*, 2013, Gotelli and Colwell, 2011)
Diversity indices	Formula	Descriptions	Reference
Simpson's Index (Yule index)	$D = \lambda = \sum_{i=1}^{S} P_i^2$	measures the probability that two individuals randomly selected from a sample will belong to the same species	(Simpson, 1949)
Simpson's Index of Diversity	$1 - D$	represents the probability that two individuals randomly selected from a sample will belong to different species	(Magurran, 2004)
Berger-Parker dominance	$d = \frac{N_{max}}{N}$	is the proportional abundance of the most abundant species	(Magurran, 2004)
Shannon index	$H' = \sum_{i=1}^{S} P_i lnP_i$	characterizes species diversity in a community	(Shannon and Weaver, 1949)
Pielou's index of evenness (J')	$E_H = \frac{H}{H_{max}} = \frac{H}{lnS}$	measures the evenness with which individuals are divided among the taxa present	(Pielou, 1975)

S = the number of species in the sample, F_1 = the number of singleton species, F_2 = the number of doubleton species.
Q_1 = the number of species that occurs in one sample only (unique species), Q_2 = the number of species that occurs in two samples.
S_{obs} = Total number of species observed in all quadrates, m= Total number of quadrates.
$P_i = \frac{n}{N}$ n_i = number of individuals of taxon ith.
N_{max} = number of individuals in the most abundant species, N = total number of individuals in the sample.

Rarefaction method

For comparing the species richness and verifying the sampling sufficiency, rarefaction curves were employed (Colwell *et al.*, 2012). This is calculated by the following formula:

$$\boldsymbol{E}(\boldsymbol{Sn}) = \sum_{i=1}^{S} [1 - \frac{\binom{N-Ni}{n}}{\binom{N}{n}}],$$

where N= total number of individuals in the sample, S= total number of species, and Ni= number of individuals of species number i.

Also, richness estimators were obtained to reduce the effects of under sampling, which inevitably biases the observed species count (Table 1).

SHE analysis

SHE analysis resolves biodiversity into three components: species richness S (as ln$S$), Shannon index H' and evenness E (as ln$E$) (Wilson *et al.*, 2012) and calculated using the following formula: $H=ln(S)+ln(E)$. SHE analysis plots show expected patterns for broken stick, log normal, and log series distributions. In the broken stick distribution, both S and H' are expected to increase and E to stay constant. The log normal distribution is associated with an increase in S and H' but a decline in E (or ln$E$/ln$S$ to stay constant) and in the log series, S and H' will remain constant, and E will decrease.

All biodiversity indices and rarefaction method were calculated using software PAST Version 3.01 (Hammer, 2013). SHE analysis and richness estimators were calculated using the software Biodiversity Pro Version 2 (McAleece *et al.*, 1997).

RESULTS

Inventory

A total of 1,231 specimens were collected; consisting of 6 genera and 6 species. The number of species collected in highland, woodland and plain areas were 6, 5 and 2, respectively (Table 2). Heydemann's classification (Table 2) and seasonal activity (Figure 1) of hard ticks in highland and plain areas showed that *Rhipicephalus bursa* (Canestrini and Fanzago, 1878) was the eudominant species with maximum activity in spring and summer, respectively. Its population has declined with the onset of autumn and winter (Figure 1). In woodland, *Boophilus annulatus* as eudominant species showed the highest activity in summer. Other dominant, subdominant or rare species usually observed in summer and autumn. For example, two rare species found in highland, are the *Ixodes ricinus* and *Dermacentor marginatus* that showed activity only in summer and autumn, respectively. It should be noted that ticks activity was near zero in winter and only one specimen of *Boophilus annulatus* was observed in woodland (Figure 1).

Species diversity components of hard ticks

The highest values of Shannon (H'=0.7) and richness (D_{Mg}=0.66; D_{Mn}=0.38) indices in highland were observed in summer while in woodland (H'=0.98) (D_{Mg}=0.56; D_{Mn}=0.51) and plain (H'=0.45) (D_{Mg}=0.56; D_{Mn}=0.82) were observed in spring (Table 3) (Figure 2). The highest values of dominance in highland (λ=0.7; d=0.82) and plain (λ=0.83; d=0.91) were observed in autumn while in woodland (λ=0.79; d=0.88) it was observed in summer (Figure 2). Evenness indices (J; $1\text{-}D$) showed the maximum values in spring, for all three topographical areas (Table 3). It should be noted that in plain the diversity indices for summer data sets could not be analyzed statistically due to the low number of species (just one species was captured). As only one specimen was captured in winter, biodiversity indices were not compared with other seasons.

Abundance of hard ticks on sheep (1011 specimens; 82.12%) is more than that on cattle (220 specimens; 17.88%). Biodiversity indices showed that the Cattle had higher richness (D_{Mn}=0.34; D_{Mg}=0.74) and diversity (H'=1.10; H_B=1.11) than sheep (D_{Mn}=0.19; D_{Mg}=0.72; H'=0.88; H_B=0.87). Evenness and equitability indices of ticks were higher for cattle (E=0.63; J=0.71) than sheep (E=0.40; J=0.49) while dominance of species for sheep

Table 2. Number of specimens captured. Abundance and dominance structure of hard ticks

Taxa	Highland			Woodland			Plain			Total areas		
	Number	Relative abundance	Dominance structure	Number	Relative abundance	Dominance structure	Number	Relative abundance	Dominance structure	Number	Relative abundance	Dominance structure
Rhipicephalus bursa	728	69.3	Eudominant	28	20	Dominant	29	72.5	Eudominant	785	63.8	Eudominant
Hyalomma detritum	180	17.1	Dominant	10	7.2	Subdominant	-	-	-	189	15.4	Dominant
Boophilus annulatus	51	4.9	Rare	67	47.9	Eudominant	-	-	-	118	9.6	Subdominant
Haemaphysalis punctata	67	6.4	Subdominant	15	10.7	Dominant	11	27.5	Dominant	94	7.6	Subdominant
Ixodes ricinus	3	0.29	Subrare	20	14.3	Dominant	-	-	-	23	1.9	Rare
Dermacentor marginatus	22	2.1	rare	-	-	-	-	-	-	22	1.8	Rare
	1051			140			40			1231		

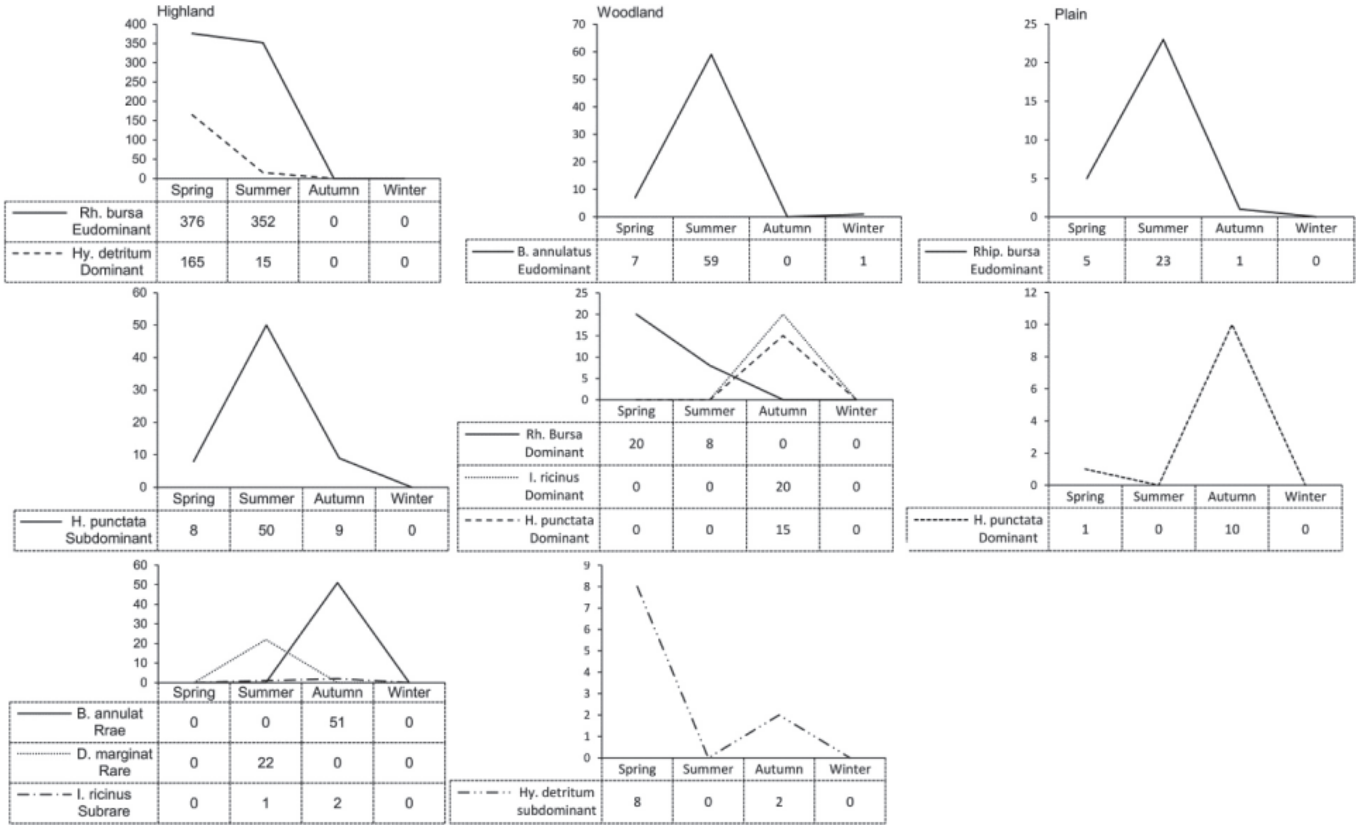

Figure 1. Seasonal dynamics of the Eudominant, Dominant, Subdominant, Rare and Subrare taxa in highland, woodland and plain areas.

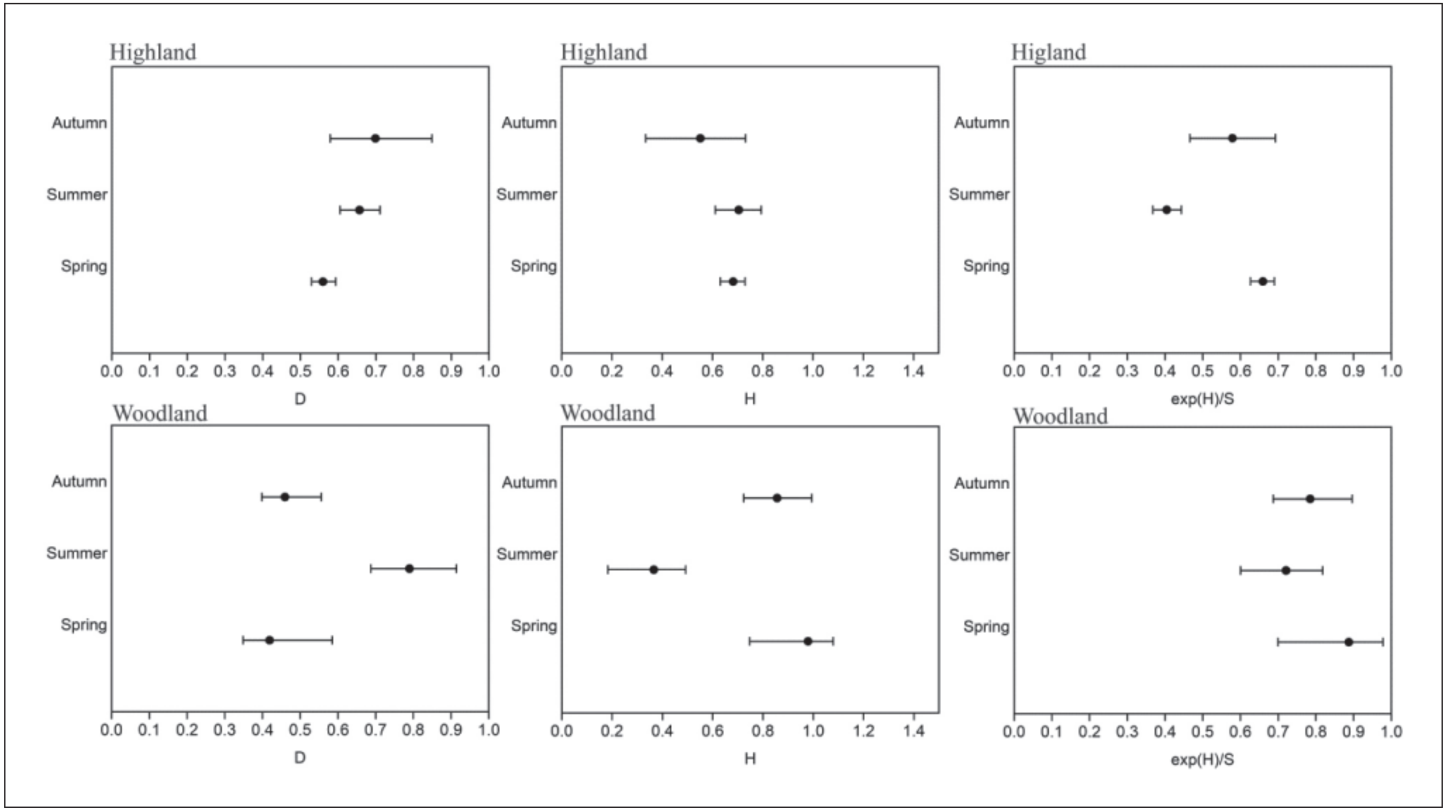

Figure 2. Biodiversity indices [Shannon (H), Dominance (D) and Evenness (exp (H/S] comparison between Highland and Woodland areas.

(D=0.59; d=0.76) was higher than that for cattle (D=0.37; d=0.45). Diversity (H' and H_B), Species richness (D_{Mn} and D_{Mg}) and evenness of female ticks was higher than males for both hosts (Table 3).

Results of diversity t-tests in highland showed that there was no significant difference between seasons (P>0.05). But in woodland, summer showed a significant difference compared to other seasons (p<0.0001) (Table 4). Also, a significant difference was observed between different hosts (p<0.0001). Finally, there was no significant difference between female and male ticks on cattle (P>0.05), but there was a significant difference on sheep (p<0.0001) (Table 4).

Rarefaction methods

Six graphs of rarefaction curves are shown in Figure 3. Three of them are based on the different seasons (left side: one for highlands, one for woodlands, and one for total areas) and the other three are based on the different hosts (right side: one for highlands, one for woodlands, and one for total seasons). Almost all rarefaction curves showed the reaching to the asymptotic line. This means that our sampling is sufficient. Only the curves of summer and autumn in highland did not reach to asymptotic line, and probably more sampling efforts are needed in these seasons.

Rarefaction curves with 95% confidence intervals (shaded areas) can compare tick richness in different seasons or hosts (Figure 3). Interpolation of sample size for summer in the highland showed that when the sample size is decreased from 440 specimens (observed richness: S_{440}=5) to 62 specimens (estimated richness: S_{62}=4.009), the richness is reduced from 5 to 4.009 (S_{440}=5, S_{62}=4.009). Interpolation of sample size for spring in the highland showed that when the sample size is decreased from 549 specimens (observed richness: S_{549}=3) to 440 (estimated richness: S_{440}=3) and 62 specimens (estimated richness: S_{62}=2.619), the richness does not change much. This implies that the estimated richness for summer in the smallest amount of sampling (S_{62}=4.009) still shows a higher value than observed richness in spring (S_{549}=3) or autumn (S_{62}=3). Similarly, in woodland, spring and autumn showed higher richness than summer (Figure 3). It should be noted that, in plain, because of the rarity of specimens and species, rarefaction curves could not be properly calculated. Finally, rarefaction curves for hosts in all topographical areas showed that the species richness on sheep is higher than cattle (Figure 3).

Richness estimators showed that estimated richness (S_{Chao1}, S_{Chao2}, S_{jack1}, S_{jack2}) was approximately equal to observed richness (S_{Obs}) (Table 5). Of course, in plain the estimated richness base on chao1 (S_{Chao1}=6.5) showed the bigger value than observed richness (S_{obs}=2) in autumn. Also, in highland estimated richness of chao1 (S_{Chao1}=7.8) and jack2 (S_{jack2}=6.8) showed the greater value than observed richness (S_{Obs}=5) in summer (Table 5).

SHE analysis

Using SHE analysis, ln(S), ln(E), and H' values were calculated cumulatively (Table 6). The SHE analysis in highland showed that the biggest values of richness (ln$S$=1.61) and diversity (H'=0.7) were observed in the summer. In woodland the biggest values of richness (ln$S$=1.1) and diversity (H'=0.98) were found in spring. In addition greater evenness in highland (ln$E$=-0.42) and woodland (ln$E$=-0.12) was observed in spring. SHE diagrams evaluated the relationships between biodiversity components (Figure 4). All measures of SHE analysis (lnS, H´, lnE/lnS and lnE) showed fluctuations of species diversity during different seasons. The SHE analysis in highland indicated that patterns of distribution in spring and autumn almost followed log normal model (increase in ln$S$ and H' but a decline in ln$E$) while distribution in summer did not follow a broken stick, log series or log normal model (Figure 4). Distribution pattern in woodland followed nearly log normal model in autumn but in spring and summer it did not follow a broken stick, log series or log normal model (Figure 4). It should be noted that because only two species were found in plain areas, SHE analysis were not calculated or plotted.

Table 3. Biodiversity indices for each seasonal and host category in all areas; separately and totally

Biodiversity Indices	Highland			Woodland			Plain			Total areas		
	Spring	Summer	Autumn	Spring	Summer	Autumn	Spring	Summer	Autumn	Spring	Summer	Autumn
Taxa (*S*)	3	5	3	3	2	3	2	1	2	4	6	5
Individuals	549	440	62	35	67	37	6	23	11	590	530	110
Dominance (*D*)	0.56	0.66	0.70	0.42	0.79	0.46	0.72	NA	0.83	0.55	0.55	0.35
Simpson (*1-D*)	0.44	0.34	0.30	0.58	0.21	0.54	0.28	NA	0.17	0.45	0.45	0.65
Shannon (*H'*)	0.68	0.70	0.55	0.98	0.37	0.86	0.45	NA	0.30	0.74	0.95	1.6
Evenness e^H/S	0.66	0.40	0.58	0.89	0.72	0.78	0.78	NA	0.68	0.52	0.43	0.64
Menhinick (D_{Mn})	0.13	0.38	0.24	0.51	0.24	0.49	0.82	NA	0.60	0.16	0.26	0.48
Margalef (D_{Mg})	0.32	0.66	0.48	0.56	0.24	0.55	0.56	NA	0.42	0.47	0.80	0.86
Equitability (*J*)	0.62	0.44	0.50	0.89	0.53	0.78	0.65	NA	0.44	0.53	0.53	0.72
Fisher alpha	0.42	0.79	0.66	0.79	0.39	0.77	1.05	NA	0.72	0.58	0.95	1.8
Berger Parker (*d*)	0.68	0.80	0.82	0.57	0.88	0.54	0.83	NA	0.91	0.68	0.72	0.46

Biodiversity Indices	Cattle			Sheep		
	Male tick	Female tick	Total (♂+♀)	Male tick	Female tick	Total (♂+♀)
Taxa (*S*)	4	5	5	4	6	6
Individuals	47	173	220	467	544	1011
Dominance (*D*)	0.58	0.40	0.37	0.63	0.57	0.59
Simpson (*1-D*)	0.42	0.60	0.63	0.37	0.43	0.41
Shannon (*H'*)	0.84	1.11	1.10	0.74	0.96	0.88
Evenness e^H/S	0.58	0.60	0.63	0.52	0.43	0.40
Menhinick (D_{Mn})	0.38	0.58	0.34	0.19	0.26	0.19
Margalef (D_{Mg})	0.78	0.78	0.74	0.49	0.79	0.72
Equitability (*J*)	0.61	0.69	0.71	0.53	0.53	0.49
Fisher	1.05	0.96	0.91	0.60	0.94	0.85
Berger Parker (*d*)	0.74	0.55	0.45	0.78	0.74	0.76

NA=not applicable

Table 4. *Student's t*-test for the significant difference in diversity (Shannon and Simpson) between different seasons and hosts

	Highlands			Woodlands			Total area		
Season	Spring-Summer	Spring-Autumn	Summer-Autumn	Spring-Summer	Spring-Autumn	Summer-Autumn	Spring-Summer	Spring-Autumn	Summer-Autumn
Shannon (H')	0.68-0.7	0.68-0.55	0.7-0.55	0.98-0.37	0.98-0.86	0.37-0.86	0.74-0.95	0.74-1.16	0.95-1.16
t-Student	0.41	1.23	1.34	5.2	0.9	4.1	3.8	6.4	2.8
P	0.7	0.2	0.2	1.3E-06	0.3	8.5E-05	0.0001	2.5E-09	0.005
Significance	NS*	NS*	NS*	Highly signif.	NS*	Highly signif.	Highly signif.	Highly signif.	Highly signif.
Simpson (D)	0.56-0.66	0.56-0.7	0.66-0.7	0.42-0.79	0.42-0.46	0.79-0.46	0.55-0.54	0.55-0.35	0.54-0.35
t-Student	3.05	1.99	6.4	4.27	0.5	4.5	0.07	6.9	5.7
P	0.002	0.051	0.5	4.7E-05	0.6	2.0E-05	0.93	3.7E-11	2.02E-08
Significance	Highly signif.	NS*	NS*	Highly signif.	NS*	Highly signif.	NS*	Highly signif.	Highly signif.

Host	Cattle-Sheep Total (♂+♀)	Cattle Female-Male	Sheep Female-Male
Shannon (H')	1.15-0.88	1.11-0.84	0.96-0.74
t-Student	4.46	1.73	3.4
P	1.02E-05	0.09	0.0008
Significance	Highly signif.	NS*	Highly signif.
Simpson (D)	0.37-0.59	0.40-0.58	0.57-0.63
t-Student	8.61	1.95	1.63
P	4.27E-17	0.06	0.1
Significance	Highly signif.	NS*	NS*

NS*: not significant (P >0.05).

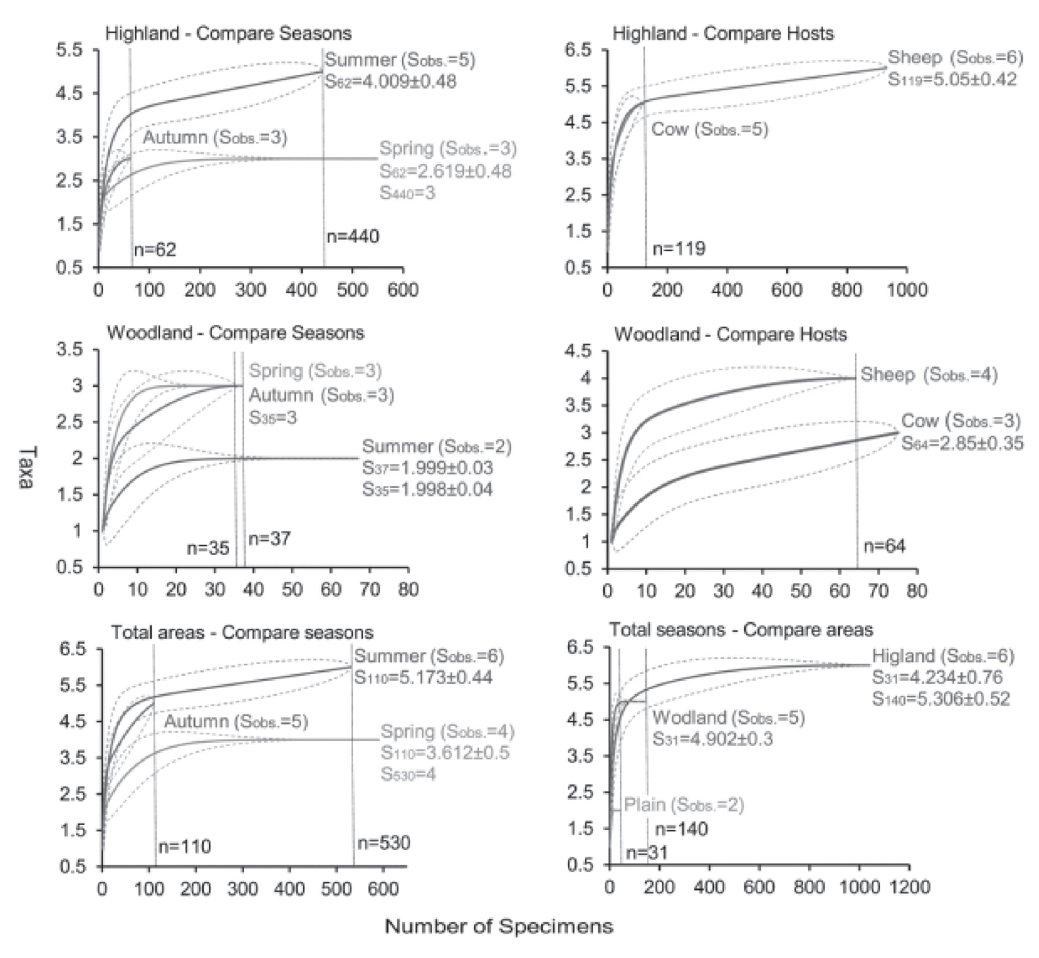

Figure 3. Individual-based rarefaction curves for species richness of hard ticks in three seasons and hosts plots in highland, woodland and plain areas with the 95% confidence limit of the rarefaction curve. The dashed vertical line indicates richness comparison standardized based on the lowest individuals. $S_{obs.}$=$S_{Observed}$, rarefied species richness is shown along with the corresponding number (S_{number}±SD).

Table 5. Total observed and estimated richness values for different seasons in Sari Township for each sampling period. Observed richness represents the number of species found in each season during each sampling period. Mean represents the arithmetic mean of the four calculated richness estimators. See Table 1 for formulae and text for description of each richness estimator

	Observed Richness	Richness Estimator Highland				Observed Richness	Richness Estimator Woodland				Observed Richness	Richness Estimator Plain			
		Chao 1	Chao 2	Jack 1	Jack 2		Chao 1	Chao 2	Jack 1	Jack 2		Chao 1	Chao 2	Jack 1	Jack 2
Season															
Spring	3	3.5	3	3	2.2	3	3.5	3.5	3.8	3.9	2	2.1	2.5	2.5	2.5
Summer	5	7.8	5	5.9	6.8	2	2	2	2.9	3.6	1	-	-	-	-
Autumn	3	3	3	3	2.6	3	3	3	3	2.4	2	6.5	2	2.8	3.4

Table 6. Results of SHE analysis data in different seasons and different areas

Sample	N	ln*N*	ln*S*	*H*	ln*E*	Ln*E*/Ln*S*
Highland						
Spring	549	3	1.1	0.68	-0.42	-0.38
Summer	440	5	1.61	0.7	-0.9	-0.56
Autumn	58	3	1.1	0.58	-0.52	-0.48
Woodland						
Spring	35	3	1.1	0.98	-0.12	-0.11
Summer	67	2	0.69	0.37	-0.33	-0.47
Autumn	37	3	1.1	0.86	-0.24	-0.22

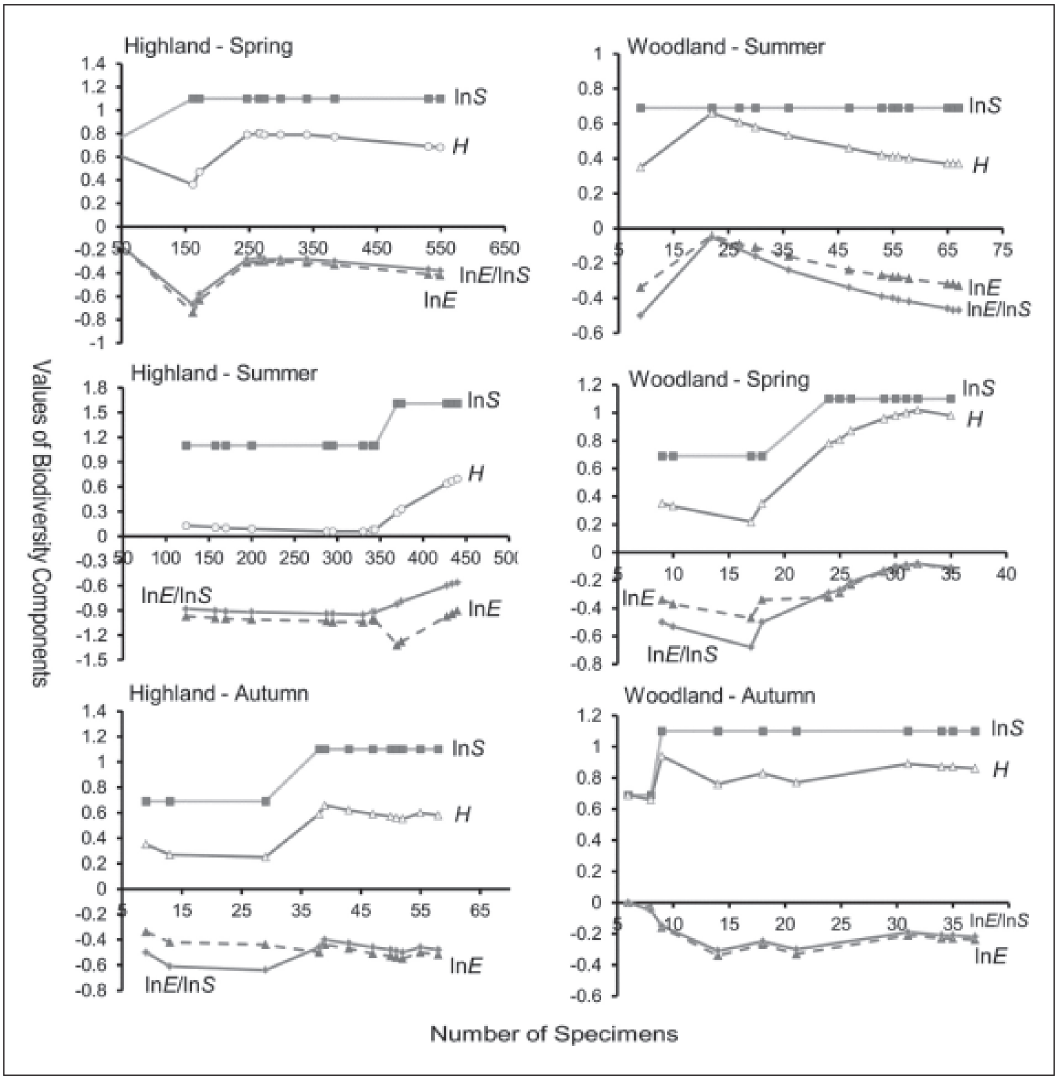

Figure 4. The results of SHE analyses on data collected in different areas and seasons. The x axis on each graph represents the accumulation species that expressed as individual cover from sample to sample. The y axis represents the diversity of richness ln(S), evenness ln(E), and Shannon H' diversity.

DISCUSSION

Rhipicephalus bursa is the most dominant tick species in the study areas, with the highest activity in spring until the middle of summer. The percentage abundance of the eudominant taxa was in accordance with that of a study in Mazandaran (Razmi *et al.*, 2007) but showed some differences to other studies (Hosseini Vasoukolaei *et al.*, 2010; Haghi *et al.*, 2013; Moghaddam *et al.*, 2014), which may be due to land exploitation, climate changes and host ecology (Rahbari *et al.*, 2007; Yakhchali *et al.*, 2011). *R. bursa* infests a wide range of domestic ruminants and is widespread in Iran, Iraq, Egypt and Turkey (Rahbari *et al.*, 2007; Akimov and Nebogatkin, 2013; Sonenshine, 1993). This species is well compatible in areas with open vegetation and different temperatures and prefers low to medium altitude, mountain, steppe and semi-desert environments (Papadopoulos *et al.*, 1996). Sari Township is situated in low-laying areas at the Caspian Sea level with a hot and humid spring and summer (Shemshad *et al.*, 2012). Furthermore, the presence of diverse vegetation in this area creates appropriate conditions for rearing a wide range of ruminants. Hence, *R. bursa* can potentially act in this area and also infest large number of livestock's populations, especially in spring and summer because this species is very active in conditions with long daylight periods and high temperature and RH. This is a very serious point because it is possible that *R. bursa* plays an important role in the transmission of *Babesia* (Razmi *et al.*, 2002; Shayan *et al.*, 2007) in different areas of Sari.

It is noteworthy that, with decreasing of the temperature, humidity and daylight, activity of *R. bursa* is sharply reduced or stopped (Gray, 2002). Therefore, with the onset of cold months the population of *R. bursa* is reduced and in opposite, the abundance of other species such as *Boophilus annulatus*, *Haemaphysalis punctata*, and *Dermacentor marginatus*, increases. It can be stated that climate change along with environmental biotic or abiotic conditions, land exploitation, host activity (Rahbari *et al.*, 2007; Yakhchali *et al.*, 2011; Shemshad *et al.*, 2012) and species competition (Thébault and Loreau; 2005Noe and Abril, 2013) are the effective factors that can play an important role about this.

For woodland and plain the highest richness and diversity were observed in spring. Usually diversity is positively related to species richness (Magurran, 2004). So, higher levels of richness in these seasons caused that the highest diversity was observed. It should be noted that although higher richness was observed on sheep but higher diversity was observed on cattle. Because, in addition to richness, evenness (positively) or dominance (negatively) can also be effective on diversity (Magurran, 2004). In this case, greater dominance and less evenness on sheep than cattle caused that the amount of diversity decreased. So, diversity was more affected by evenness or dominance than richness. Finally, female ticks had more abundance, richness, evenness and diversity than male; an observation that was in accord with other studies (Razmi *et al.*, 2007; Shemshad *et al.*, 2012).

Base on rarefaction curves for woodland and highland the maximum richness was observed in spring and summer, respectively (Figure 3). These curves showed that our sampling effort provided maximum richness because species richness reached to the asymptotic line (Stireman III *et al.*, 2014). Therefore, sampling efforts were enough in our study; it means that with additional sampling effort more new species could not be found (Dove and Cribb, 2006; Stireman III *et al.*, 2014; Nikookar *et al.*, 2015). In this regard, species richness estimators approximately confirmed that our sampling effort provided a good representation of hard tick richness.

SHE analysis for highland elucidated that in spring the diversity is sensitive to evenness, so that with decreasing or increasing of evenness the diversity is also decreased or increased, respectively. Fluctuations of diversity components (H', ln$S$ and lnE/lnS) in spring confirmed that tick distribution did not follow from any particular pattern and so, species are relatively common (Small and McCarthy, 2002; Murray, 2003). Summer diagram for highland showed that ln(*E*)/ln(*S*) and *H´* were comparatively constant and, hence, tick community possibly exhibited characteristics between log normal and log series distributions (Small and McCarthy, 2002). It means that the community had a small number of dominant species (Buzas and Hayek, 2005; Shott, 2010), such as *R. bursa*, and a relatively large proportion of rare species (Buzas and Hayek, 1996; Small and McCarthy, 2002), such as *B. annulatus*, *D. marginatum* and *I. ricinus*. Autumn diagram for highland showed that tick diversity is sensitive to richness. Also, ln(*E*)/ln(*S*) remained moderately constant and consequently log normal was the best pattern for distribution. This kind of distribution indicated that ticks community had a proportion of rare species (Buzas and Hayek, 1996). SHE diagrams for woodland showed that tick diversity was sensitive to richness and also tick distribution did not follow any distribution pattern.

CONCLUSION

It can be stated that our study provides a baseline survey of noticeable fluctuations of diversity components in different spatial and temporal scales. Our findings indicated that richness and evenness had a different impact on tick's diversity. In this context, rarefaction and SHE analysis were consequently allowed to standardize samples for meaningful comparisons. Our results suggested that hard tick communities comparatively contained common species. But probably more sampling efforts may be necessary for summer and autumn to adequately evaluate richness, especially in highland and plain. Finally, we determined that in spring and summer, changes in dominant species such as *R. bursa* cause sharp fluctuations in evenness and therefore, may increase the rates of exposure to tick-borne diseases.

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