

Rodent species diversity and occurrence of *Leishmania* in northeastern Iran

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ABSTRACT

The aim of the current study was to survey the species diversity and *Leishmania* infection of rodents in the Gonbad -e- Kavoos City and County, northeastern Iran, during 2018–2019. Totally, 20 rural and four urban areas endemic for leishmaniasis were selected and visited monthly. Rodent species diversity indices were calculated in different spatial and temporal scales. Species richness was also calculated using rarefaction curves to estimate the number of species in unequal sample sizes. A total of 167 rodents belonging to 7 species, 6 genera, and 2 families were collected. *Rattus rattus* (34.7%) and *Rattus norvegicus* (18.6%) were identified as dominant species. The largest Margalef and Menhinick species richness ($D_{Mg} = 1.55$; $D_{Mn} = 1.01$) and the highest value of the Shannon index ($H' = 1.71$) were observed in rural areas. Seasonal abundance of the rodents varied among species and showed that the highest richness ($D_{Mg} = 1.27$; $D_{Mn} = 0.65$) occurred in spring and the highest value of Shannon index ($H' = 1.72$) was noted in summer. Rarefaction curves confirmed that the highest species richness occurred in summer and spring in the rural area. Seasonal fluctuations of *R. rattus* and *R. norvegicus* have a significant impact on the changes in rodent species diversity. Investigations of *Leishmania* infection showed that amastigote forms of *Leishmania* were observed in 15.4% of smears taken from *Rhombomys opimus* and *Meriones libycus* and examined under the light microscope. The highest rate of *Leishmania* infection was observed during the summer.

INTRODUCTION

Rodents are small-sized animals forming the most numerous group of mammals (Canales-Brellenthin 2020). Until now, over 2200 species of rodents have been identified; that ap-

proximately comprise 42% of the world's total mammalian diversity (Wilson and Reeder 2005). Hundreds of studies on rodent fauna have been performed worldwide which have increased our knowledge about distributional patterns of rodents (Du *et al.* 2017, Putnam

et al. 2018). Rodents live in different habitats and play an important role in public health and the human economy. For example, in the United States, the economic cost of rodents' damages was estimated at more than \$19 billion every year (Pimentel *et al.* 2000). In the field of public health, rodents also play an important role as hosts or reservoirs for many ecto- and endoparasites.

Iran is located in the Palearctic region (32°42'79"N, 53°68'80"E) and comprises a variety of geographic and climatic habitats with a high variety of rodent species in different areas. More than 79 rodent species have been recorded from Iran which is almost one-third of all mammal species of the country (Karimi *et al.* 2008). Given ubiquity of rodents in different parts of Iran, rodent species diversity is poorly investigated. Nevertheless, study on rodent species diversity can provide important information about ecosystem conditions, environmental changes or ecological disturbance (Darvish *et al.* 2006, Ghorbani *et al.* 2014, Amirafzali *et al.* 2017, Esfandiari *et al.* 2017, Arzamani *et al.* 2018, Shad and Darvish 2018). Therefore, it seems very important to assess rodent species diversity in Iran.

In Iran, as well as many parts of the world, rodents are known as important reservoirs of infectious diseases in many provinces (Esfandiari *et al.* 2015). One of the most common of these diseases is leishmaniasis, so that people in 18 out of 32 provinces across the country are at risk of contracting the disease (Mohebbali *et al.* 2004). This disease, which is caused by the *Leishmania* parasite, occurs in both cutaneous (in the north, east and south) and visceral (sporadically) forms in the country (Mohebbali *et al.* 2004). In Iran, *Leishmania* parasite uses different species of rodents as reservoirs including *Rhombomys opimus*, *Meriones libycus*, *Meriones hurrianae*, *Tatera indica* (cutaneous leishmaniasis), *Cricetulus migratorius*, *Mesocricetus auratus*, and *Meriones persicus* (visceral leishmaniasis); two of them *R. opimus*, and *M. libycus* are the most important hosts in the northern regions of Iran (Mohebbali *et al.* 2004), where the study was conducted.

Rhombomys opimus is reported from north-east (MinooDasht district) and central (Badrood district, Ardakan district, Sabzevar district) regions of Iran and *Meriones libycus* is reported from north-east (MinooDasht dis-

trict), south-west (Fars province) and central areas (Ardakan district) of Iran (Mohebbali *et al.* 2004). *R. opimus* and *M. libycus*, which are usually infected by *Leishmania major*, play a very important role in the spread of cutaneous leishmaniasis in endemic areas of Iran. The study of the frequency of *Leishmania* infection in the smears from *R. opimus* and *M. libycus* in several endemic areas of Iran showed that *R. opimus* is the principal reservoir of cutaneous leishmaniasis in the north-eastern Iran with 85.2% infection rate. This study also showed different frequencies of *Leishmania* infection in this species in the central parts of Iran, from 36.4% (Sabzevar district) and 32.0% (Badrood district) to 11.5% (Ardakan district). Also, *M. libycus* showed different frequency of *Leishmania* infection from 35.1% (Fars province) and 25.0% (Badrood district) to 15.8% (Ardakan district) of infection rate in the central and south-west areas of Iran (Mohebbali *et al.* 2004).

There are several foci of leishmaniasis, especially cutaneous form, in the north, east and south of Iran. One of the important and high-risk areas for leishmaniasis in northeastern Iran is Golestan Province (37°28'98"N, 55°13'76"E) (Mollalo *et al.* 2015). Gonbad -e- Kavoods County being one of the main foci of cutaneous leishmaniasis in Iran is located in the northern and central part of Golestan Province with an elevation of 38 m above sea level. About 4% of the population of Gonbad -e- Kavoods suffers from acute ulcer and 78% from scar of cutaneous leishmaniasis caused by *Leishmania* parasite (Sofizadeh *et al.* 2012). In this regard, there are two species of rodents: *R. opimus* and *M. libycus* that play a key role as reservoirs of *Leishmania* parasite in this County (Sofizadeh *et al.* 2016). Gonbad -e- Kavoods has approximately 150 km of common border with the Republic of Turkmenistan with mainly desert and semi-desert environment. This is very important issue because on the one hand leishmaniasis is endemic in these areas and on the other hand 15 species of rodents were identified and reported from border areas of northeastern Iran and Turkmenistan (Ponirovskii and Darchenkova 2008, Arzamani *et al.* 2018), therefore these areas can be active pathways in the transmission of the disease via the travel of infected people and rodent migration.

Given the importance of these issues and poor knowledge of the local rodent species diversity, the survey of fauna and species diversity of rodents in this region can be an important ecological and medical subject. Therefore, the main objective of this study was to investigate the community of rodents from different habitats within urban and rural areas to determine the species diversity parameters (richness, Shannon index, and dominance index), abundance, and seasonal dynamics of rodents during 12 months. Another objective of this study was to screen *Leishmania* infection in *R. opimus* and *M. libycus*, considered as *Leishmania* reservoirs, and their possible role in the transmission of the disease to human in Gonbad -e- Kavoods.

MATERIALS AND METHODS

Study area

This study was conducted during 2018–2019 in Gonbad -e- Kavoods City and County (37°24'07"N, 55°15'97"E), an enzootic cutaneous leishmaniasis focus in northeastern Iran, which is bordered to the north by the republic of Turkmenistan. The area has a mild and humid climate, and the annual average of temperature, precipitation, and relative humidity is 18.6°C, 500 mm and 65%, respectively. We studied the species diversity of rodent assemblages in 24 sites located in 20 rural areas and four urban areas of Gonbad -e- Kavoods (Fig. 1, Appendix 1). Since the rate of leishmaniasis in

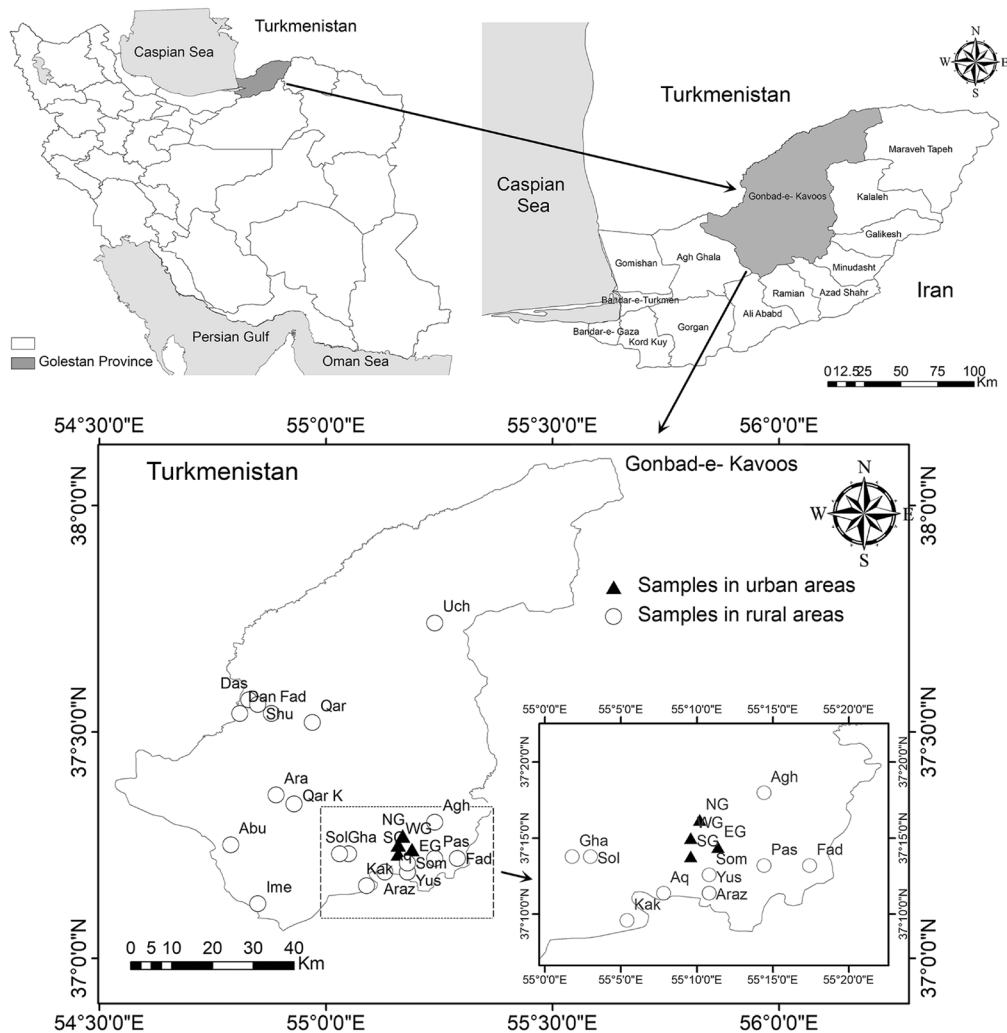


Fig. 1. Geographical locations of sampling sites of the rodents in Gonbad -e- Kavoods City and County, Iran, 2018–2019. Abbreviations indicate sampling sites. For the full names and more information see Appendix 1.

the urban area is known to be homogeneous, four sampling sites in the north, south, east and west were selected to cover the entire urban area of Gonbad -e- Kavoos. Sampling sites in rural areas were selected based on knowledge of core areas of the disease. For this purpose, all reports of leishmaniasis cases received from various rural health centers were studied and then 20 rural areas with the highest disease rate were selected and sampled (Fig. 1).

Rodent trapping and collection

Rodent sampling started in April 2018 and ended in March 2019 covering all seasons. Specimen collection was performed in accordance with the procedures approved by the Ethical Committee of Mazandaran University of Medical Sciences. Rodent trapping was carried out in 4 urban areas (20 sampling sites) and 20 rural areas (100 sampling sites). Based on the study design, at each sampling site, 5 locations (one in the center and four in different geographical directions from the center with a distance of 200 to 400 meters) were selected for trapping and 5 traps were placed in each location. Rodent trapping was carried out by five teams in different sampling sites rotationally. For rodent collection, the burrows were considered active if they showed signs such as the presence of fresh droppings, hemp of soil, trodden pathways around the holes, and absence of spider web around the holes (Mohammadi *et al.* 2020). The samples were collected using Sherman live traps. All traps were set at sunset or sunrise for trapping nocturnal or diurnal species, respectively. A mixture of peanut butter with bread and a variety of fruit and vegetables, depending on the season, were used as bait to attract the rodents. All traps were monitored every morning for nocturnal species and every evening for diurnal species (Mohammadi *et al.* 2020). Trapping was done twice a month within different habitats or vegetation types from April 2018 to March 2019. After transferring the rodent to the laboratory, standard measurements of morphological traits (such as weight, sex of rodents, tail length, length of head and body, ear length, rear foot size) were recorded and then the rodents were identified to the species using appropriate keys (Corbet 1978, Etemad 1979).

Investigation of *Leishmania* infection

The main objective of this study was to screen rodents for *Leishmania* infection for a better understanding of the epidemiology of the disease in Gonbad -e- Kavoos. For the leishmanial infection assay, the captured rodents were transferred to the laboratory and were anesthetized with xylazine-ketamine. The total samples of 29 *Rhombomys opimus* and 10 *Meriones libycus* were examined for *Leishmania*. Smears were taken from the skin of the ears and snout of each rodent. The smears were fixed in methanol, stained by Giemsa, and were examined for parasites by light microscopy using high magnification ($\times 100$). The samples from infected rodents were cultured in Novy-MacNeal-Nicolle (NNN) and RPMI-1640 culture. The cultures were checked for promastigotes twice a week for a period of 6 weeks.

Data analysis

Species richness, dominance and diversity were evaluated using:

Margalef index

$$(D_{Mg} = \frac{S-1}{\ln N})$$

Menhinick index

$$D_{Mn} = \frac{S}{\sqrt{N}}$$

Simpson's dominance index

$$(D = \lambda = \sum_{i=1}^S P_i^2)$$

and Shannon index ($H' = -\sum p_i \times \ln p_i$). To compute whether species were dispersed uniformly, Pielou's index of evenness ($E = \exp(H')/S$) was used (Shannon and Weaver 1949, Magurran 2004, Magurran *et al.* 2010). In the above equations, N is the total number of individuals in the sample, S is the number of species in the sample,

$$P_i = \frac{n_i}{N}$$

where P_i is the proportion of individuals found in the i th species, and n_i is the number of individuals of the taxon i th. Diversity test was then performed by comparing the Shannon and Simpson diversities in two samples

(Jacobson *et al.* 2011). To verify the sampling sufficiency to assess true richness and to determine the observed and estimated richness of rodents, rarefaction curves were applied using the following formula:

$$E(Sn) = \sum_{i=1}^s \left[1 - \frac{\binom{N-N_i}{n}}{\binom{N}{n}} \right]$$

where N = total number of individuals in the sample, S = total number of species, and N_i = number of individuals of species number i (Chao *et al.* 2014).

RESULTS

Rodent abundance and seasonality

During this study, a total of 167 rodents were captured. Seven species belonging to two families of Muridae and Dipodidae were recorded in the following proportions: 34.7% *Rattus rattus*, 18.6% *Rattus norvegicus*, 17.4% *Rhombomys opimus*, 12% *Mus musculus*, 9% *Pygeretmus pumilio*, 6% *Meriones libycus* and 2.4% *Allactaga elater* (Appendix 2). The analysis of the seasonality of rodent occurrence

showed that *R. rattus* and *R. norvegicus* were captured in all months of the year (Fig. 2).

In contrast, *A. elater* was caught only in June and July. Other species were captured in different months. It should be noted that the highest number of species (S) and abundance (n) were observed in June ($S = 7$; $n = 24$) and July ($S = 7$; $n = 23$) (Fig. 2; Appendix 3).

The rodents' spatial distribution is shown in Figure 3. Results showed that *R. rattus* ($n = 58$) was the most common, occurring in 14 different areas of the study (58.3%); next was *R. norvegicus* ($n = 31$) found in 9 areas (37.5%); and *Mus musculus* ($n = 20$; 8 areas; 33.3%). The other 4 species were somewhat less common: *R. opimus* ($n = 29$; 5 areas; 20.8%), *P. pumilio* ($n = 15$; 3 areas; 12.5%), *M. libycus* ($n = 10$; 3 areas; 12.5%) and *A. elater* ($n = 4$; 1 areas; 4.2%). It should be noted that *R. opimus*, *P. pumilio*, *M. libycus* and *A. elater* were only captured in rural areas and *R. rattus*, *R. norvegicus* and *M. musculus* were caught in both urban and rural areas (Fig. 3).

Rodent species diversity

Analysis of rodent species diversity indices in the spatial scale indicated that the highest species richness occurred in the rural areas

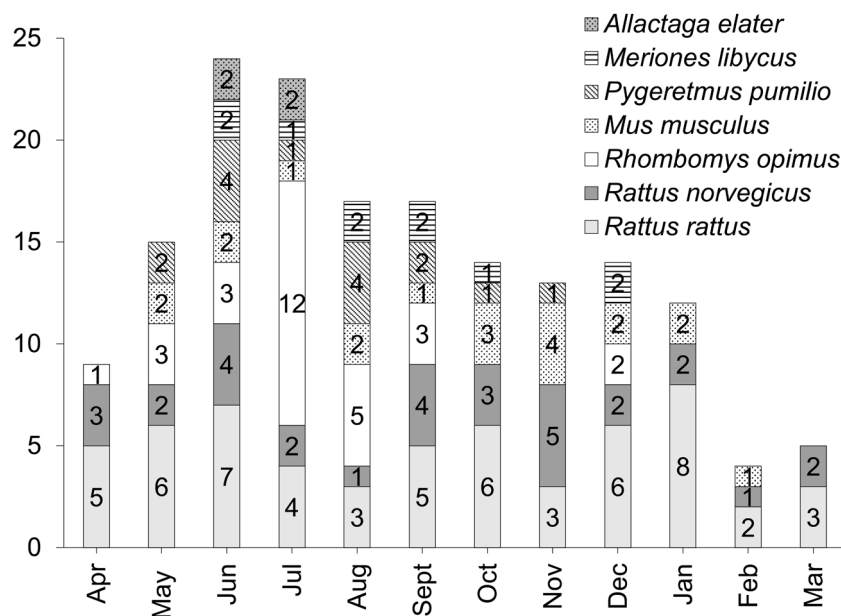


Fig. 2. Species composition of rodents trapped in enzootic cutaneous leishmaniasis foci in the northeastern Iran in different months between April 2018 to March 2019. The numbers inside each column indicates the number of captured individuals of each species in a given month in all sampling sites.

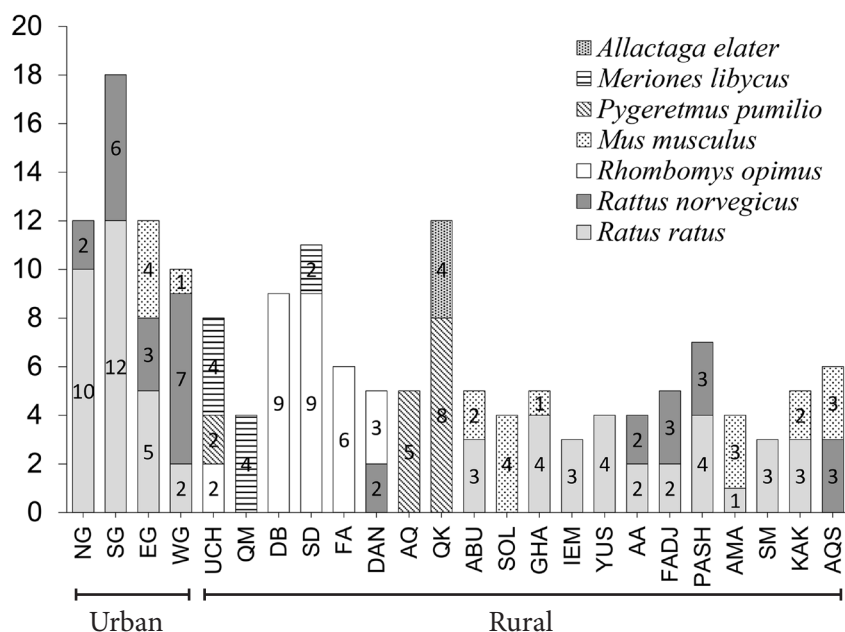


Fig. 3. Species composition of rodents trapped in twenty-four enzootic cutaneous leishmaniasis foci in northeastern Iran. The numbers inside each column indicate the number of captured individuals of each species. Abbreviations indicate sampling sites. For more information see Appendix 1.

Table 1. Abundance (n, %) and species diversity, richness, and evenness indices (H' , D , D_{Mg} , D_{Mn} , E) of rodents collected in different seasons and areas in the Gonbad -e- Kavoos (northeastern Iran).

Species	Season				Area	
	Spring	Summer	Autumn	Winter	Urban	Rural
Black rat <i>Ratus ratus</i>	18 (31)	12 (21)	15 (26)	13 (22)	29 (50)	29 (50)
Brown rat <i>Rattus norvegicus</i>	9 (29)	7 (23)	10 (32)	5 (16)	20 (64.5)	11 (35.5)
Great gerbil <i>Rhombomys opimus</i>	7 (24)	20 (69)	2 (7)	-	-	29 (100)
House mouse <i>Mus musculus</i>	4 (20)	4 (20)	9 (45)	3 (15)	5 (25)	15 (75)
Fat-tailed jarboa <i>Pygeretmus pumilio</i>	6 (40)	7 (47)	2 (13)	-	-	15 (100)
Libyan jird <i>Meriones libycus</i>	2 (20)	5 (50)	3 (30)	-	-	10 (100)
Small five-toed jarboa <i>Allactaga elater</i>	2 (50)	2 (50)	-	-	-	4 (100)
Total	48 (28.7)	57 (34.1)	41 (24.6)	21 (12.6)	54 (32.3)	113 (67.7)
Species richness and diversity indices						
Shannon Index, H'	1.69	1.72	1.53	0.91	0.91	1.71
Simpson Index, D	0.22	0.21	0.25	0.46	0.44	0.2
Margalef Index, D_{Mg}	1.55	1.48	1.34	0.65	0.50	1.27
Menhinick Index, D_{Mn}	1.01	0.92	0.93	0.65	0.40	0.65
Evenness, E	0.77	0.80	0.77	0.83	0.83	0.79

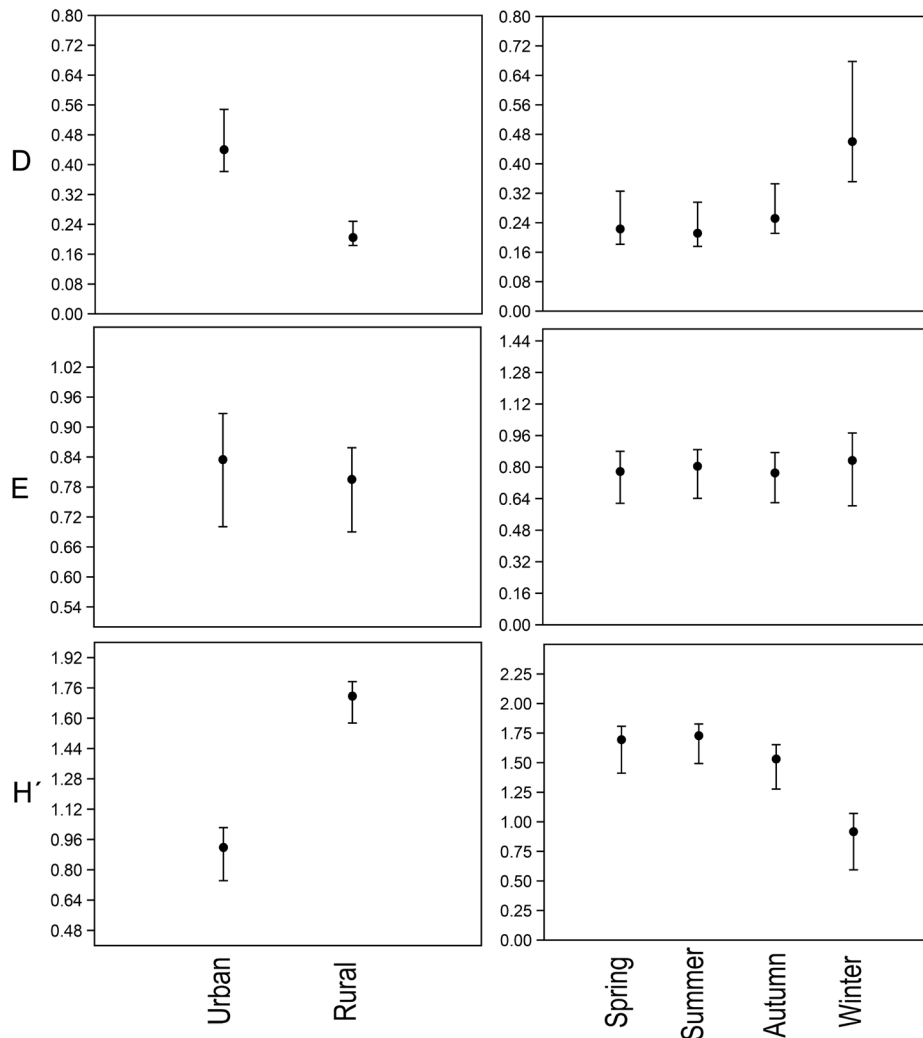


Fig. 4. Comparison of rodent diversity indices (Dominance, D, Evenness, E and Shannon, H') among different spatial (region) and temporal (season) scales in Gonbad -e- Kavvoos, northeastern Iran. Mean values \pm SE are presented.

while the urban areas had the lower values of the richness index (Table 1, Fig. 4). Evenness and Dominance indices of rodents were higher for urban areas than rural areas, while the Shannon index was significantly ($P < 0.05$) higher in rural areas than in urban areas. Species richness in different seasons showed the highest value in spring. Evenness and dominance indices of rodents were higher in winter than in other seasons while the Shannon index showed the highest species diversity in summer. The Shannon and Dominance indices showed the significant differences between winter and other seasons ($P < 0.05$) (Table 1, Fig. 4).

Rarefaction curves were constructed to evaluate the asymptotic numbers of species and their similarity between different seasons and areas (Nikookar *et al.* 2015, Fazeli-Dinan *et al.* 2019). Individual-based rarefaction curves for species richness of rodents in different seasons and areas indicated that for the given sample size, the highest richness occurred in spring and summer as well as in rural areas. In the current study, the rarefaction plots for different months and areas presented flattened asymptotic curves, thus, proving that the sampling effort was sufficient in all months and areas to register the maximum number of rodent species present (Fig. 5).

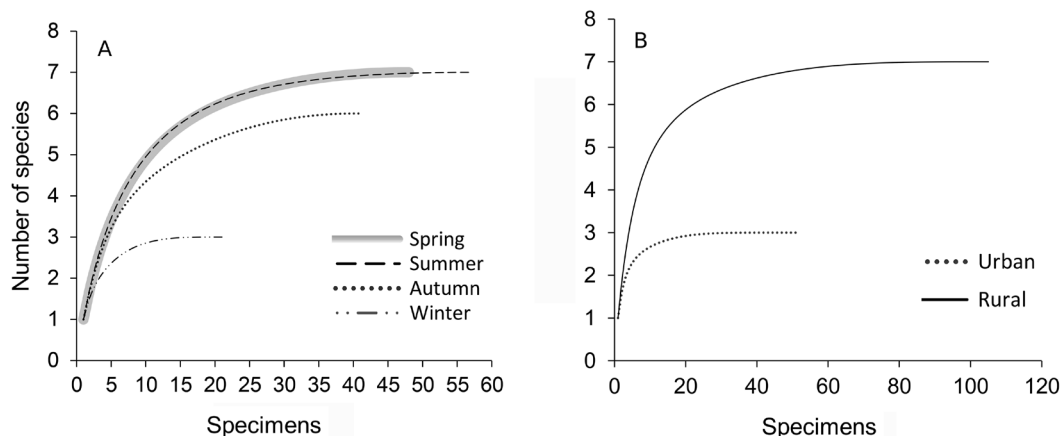


Fig. 5. Individual-based rarefaction curves for species richness of rodents in different seasons (A) and areas (B) in northeastern Iran.

Rodent infection by *Leishmania*

Leishmania amastigotes were observed in the smears taken from 6 (15.4%) rodents by direct microscopy examination. The highest rate of *Leishmania* infection was observed in rodents collected during summer (Table 2).

DISCUSSION

The studies of rodent species diversity and distribution are important because rodents are linked to human health and economics (Darvish *et al.* 2014). In this study, *R. rattus* as a dominant species was present in all seasons. This species is highly adaptable to different habitats and harms human health and the economy. *R. rattus*, although not known as *Leishmania* reservoir, can transmit at least twenty different pathogenic and infectious agents such as *Escherichia coli*, *Leptospira* spp., *Hymenolepis diminuta*, *H. nana*, *Capillaria hepatica* and *Toxoplasma gondii* (Almeida *et al.* 2013, Clapperton *et al.* 2019, Rehman *et al.* 2019, Strand and Lundkvist 2019); therefore its presence in the region as a dominant species is not desirable. In this study, *A. elater* and *P. pumilio* were rare, present only in areas on the altitude of less than 25 m above sea level. The presence of these two species depends largely on vegetation, such as *Artemisietum* spp., *Halostachys* spp., and *Haliidium* spp.; as well as suitable

soil type (Colak and Yigit 1998, Moradi and Moradi 2013).

The results indicated that *R. opimus* (April to September) and *M. libycus* are mostly present in warm months (June to October). Previous studies suggested that climatology and topography variables such as temperature, seasonal rainfall, height and slope of the area have a great impact on the distribution and consequently capture of *R. opimus* and *M. libycus* species (Gholamrezaei *et al.* 2016). These two species which are generally native to central Asia, Iran, Afghanistan and other neighboring countries (Ghatee *et al.* 2020), are usually found in rural and suburban areas less than 500 meters above sea level (Ghorbani *et al.* 2014) which is consistent with the location of our study sites. *R. opimus* and *M. libycus* are two medically important rodents as *Leishmania* reservoirs and play a key role in maintaining zoonotic cutaneous leishmaniasis in rural areas (Sofizadeh *et al.* 2016). In the current study, the leishmanial infection was observed in these two species which confirms their role as the hosts for leishmaniasis in the study area. This finding is important because reports indicate that the incidence rate for zoonotic cutaneous leishmaniasis in Gonbad -e- Kavous in the Golestan Province is higher than in other counties (Sofizadeh *et al.* 2018). Our results also indicated that the higher rate of *Leishmania* infection was observed in *R. opimus* than in *M. libycus*, therefore the former species is identified here

Table 2. *Leishmania* infection of rodents caught in rural areas of the Gonbad -e- Kavoods across the seasons of the 2018–2019. *R. opimus* – *Rhombomys opimus*, *M. libycus* – *Meriones libycus*. For site full names see Appendix 1.

Sampling site	Rodent species	Total number tested (total positive No. on microscopy)	Number tested in each season (positive No. on microscopy)				<i>Leishmania</i> species identified
			Spring	Summer	Autumn	Winter	
Uch	<i>R. opimus</i>	2 (-)	-	1(-)	1(-)	-	-
	<i>M. libycus</i>	4 (1)	-	2(1)	2(-)	-	<i>L. major</i>
Dan	<i>R. opimus</i>	3 (2)	1(1)	2(1)	-	-	<i>L. major</i>
	<i>M. libycus</i>	NC	-	-	-	-	-
Das	<i>R. opimus</i>	9 (2)	1(1)	8(1)	-	-	<i>L. major</i>
	<i>M. libycus</i>	NC	-	-	-	-	-
Shu	<i>R. opimus</i>	9 (1)	2(-)	7 (1)	-	-	<i>L. major</i>
	<i>M. libycus</i>	2 (-)	-	1(-)	1(-)	-	-
Fad	<i>R. opimus</i>	6 (-)	3(-)	2(-)	1(-)	-	-
	<i>M. libycus</i>	NC	-	-	-	-	-
Qar	<i>R. opimus</i>	NC	-	-	-	-	-
	<i>M. libycus</i>	4 (-)	2(-)	2(-)	-	-	-

NC: Indicates that the species (*R. opimus* or *M. libycus*) was not caught in respective area.

as the main reservoir host for zoonotic cutaneous leishmaniasis in this area. This species which is distributed also in other provinces of Iran such as Golestan, Khorassan-e-Shomali, Khorassane-Razavi, Khorassan-e-Jonoobi, Yazd, Isfahan, Semnan, Kerman and Sistanva-Baluchestan, has a widespread distribution and high infection rate for *Leishmania major*, thus it has high potential for spreading leishmaniasis (Sofizadeh *et al.* 2018). Therefore, it is recommended that the Ministry of Health carries out the control program to reduce this reservoir population and lower the leishmaniasis incidence in the study areas.

In the current study, rarefaction curves were asymptotic; therefore further sampling effort per month or area would not increase the evaluation of the rodent species richness (Duelli *et al.* 1999, Nikookar *et al.* 2015). Standard rarefaction analysis showed also that species richness of rodents was lower in urban areas. In addition, a decrease in the Shannon index and consequently a decrease in rodent species diversity were also observed in urban areas, which is related to human activities in urban areas. Human activities and urbanization usually destroy rodent natural habitats,

food resources and even native predators of rodents and, consequently, have a negative impact on their presence and species diversity (Guevara and Ball 2018). Decreasing rodent species diversity in urban areas can be also caused by the increase in populations of dominant species such as *R. rattus* and *R. norvegicus*, which are well adapted to human environments. Our results are consistent with those of other studies which suggest that urban areas are generally characterized by lower richness and diversity of species than natural ecosystems and also have a high proportion of invading rodents such as *R. rattus* and *R. norvegicus* (Fernández and Simonetti 2013, Byers *et al.* 2019, Strand and Lundkvist 2019). In this study, rodent species diversity components such as richness and Shannon indices were higher during spring and summer than in autumn and winter which may be explain by worsening environmental conditions and lower food availability for rodents (Brown and Ernest 2002, McClelland *et al.* 2018).

In conclusion, it can be stated that our study provides a baseline survey of variation in rodent diversity in different spatial and temporal scales. Our findings indicated that

urbanization may not be able to support viable populations of native rodents and by eliminating native rodents dramatically decreases species richness and rodent species diversity. It is possible that the reduction in richness, evenness and species diversity in urban areas and/or cold seasons could mostly be due to the dominance of *R. rattus* and *R. norvegicus*, habitat loss and destruction of plant communities in urban areas. Also, since *R. opimus* and *M. libycus*, as reservoirs of *Leishmania* parasite were observed only in rural areas, this study largely indicated that continuous surveillance on rodents is needed in the rural areas of the Gonbad -e- Kavoods to control the population of these two reservoir species.

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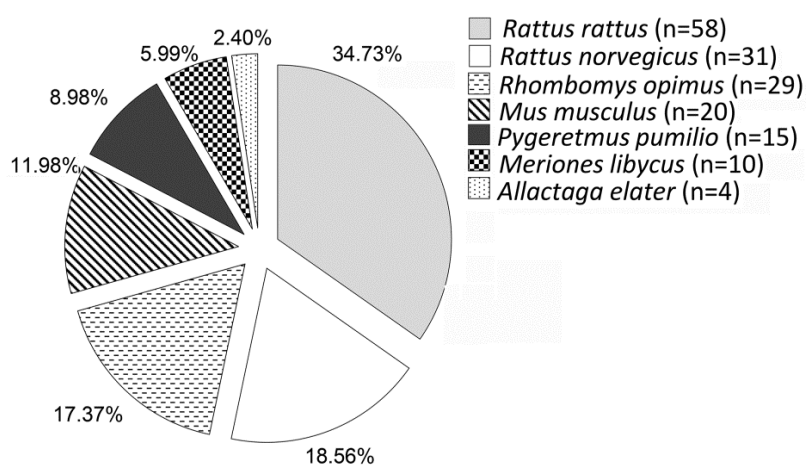
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Appendix 1. Characteristics of sampling sites of the rodents in Gonbad -e- Kavous City and County, Iran, 2018–2019.

Area	Sampling site (local name)	Site abbreviation	Latitude (N)	Longitude (E)	Above sea level (m)
Urban	North of Gonbad -e- Kavus	NG	37.27	55.17	40
	South of Gonbad -e- Kavus	SG	37.23	55.16	37
	East of Gonbad -e- Kavus	EG	37.24	55.19	42
	West of Gonbad -e- Kavus	WG	37.25	55.16	38

Area	Sampling site (local name)	Site abbreviation	Latitude (N)	Longitude (E)	Above sea level (m)
Rural	Uchquyi	Uch	37.74	55.24	185
	Qareh makher	Qar	37.52	54.97	44
	Dashli Brun	Das	37.57	54.83	27
	Shur Degesh	Shu	37.61	54.83	23
	Fadavi	Fad	37.63	54.81	46
	Daneshmand	Dan	37.50	54.78	26
	Ara Qui	Ara	37.40	54.89	22
	Qarah Kasalkheh	QarK	37.34	54.93	23
	Abuzer	Abu	37.33	55.40	102
	Soltanali	Sol	37.23	55.05	29
	Ghazaghli	Gha	37.23	55.03	28
	Imer-e-molāsāri	Ime	37.12	54.85	20
	Yusefabad	Yus	37.41	55.38	101
	Agh Abad	Agh	37.30	55.24	50
	Fadjr	Fadj	37.51	55.50	151
	Pashmak	Pas	37.16	55.19	69
	Araz Mohammad Akhund	Araz	37.54	55.56	157
	Someh Makhtūm	Som	37.21	55.18	43
	Kaka	Kak	37.16	55.09	54
	Āq Chali-ye Soflá	Aq	37.17	55.15	52

Appendix 2. Species compositions of rodents trapped at enzootic cutaneous leishmaniasis foci in northern Iran in all studied areas (n; collection number).



Appendix 3. Distribution of rodent species in four seasons according to the different locations (*: occurrence status; NC: not captured). Abbreviations indicate sampling sites. For more information see Appendix 1.

Season	Species	Urban				Rural																Presence %						
		NG (2)‡	SG (2)	EG (3)	WG (3)	Uch (3)	Qar (1)	Das (1)	Shu (2)	Fad (1)	Dan (2)	Ara (1)	Qar (2)	Abu (2)	Sol (1)	Gha (2)	Ime (1)	Yus (2)	Agh (2)	Fadj (2)	Pas (2)		Araz (2)	Som (1)	Kak (2)	Aq (2)		
Spring	<i>R. rattus</i>	*	*	*	*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	41.7	
	<i>R. norvegicus</i>	*	*	*	*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	29.2	
	<i>R. opimus</i>	-	-	-	-	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	16.7	
	<i>Mu. musculus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	16.7	
	<i>P. pumilio</i>	-	-	-	-	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	12.5
	<i>M. libycus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4.2	
Summer	<i>A. elater</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4.2	
	<i>R. rattus</i>	*	*	*	*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	29.2	
	<i>R. norvegicus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	20.8	
	<i>R. opimus</i>	-	-	-	-	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	20.8	
	<i>M. musculus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	12.5	
	<i>P. pumilio</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	12.5	
Autumn	<i>M. libycus</i>	-	-	-	-	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	12.5	
	<i>A. elater</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4.2	
	<i>R. rattus</i>	*	*	*	*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	33.3	
	<i>R. norvegicus</i>	*	*	*	*	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	20.8	
	<i>R. opimus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	8.3	
	<i>M. musculus</i>	-	-	-	-	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	29.2	
Winter	<i>P. pumilio</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	8.3	
	<i>M. libycus</i>	-	-	-	-	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	8.3	
	<i>A. elater</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	NC		
	<i>R. rattus</i>	-	-	-	-	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	33.3	
	<i>R. norvegicus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	16.7	
	<i>R. opimus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	NC	
<i>M. musculus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	8.3		
<i>P. pumilio</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	NC		
<i>M. libycus</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	NC		
<i>A. elater</i>	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	NC	

‡The numbers in parentheses indicate the number of species caught in each area.