Original Article

Biodiversity and Spatial Distribution of Mosquitoes (Diptera: Culicidae) in Kurdistan Province, Western Iran

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Abstract

Background: Mosquitoes (Diptera: Culicidae) have always been considered as the vector/s of viral and parasitic diseases. This study aimed to conduct a comprehensive survey on the species composition, spatial distribution, and biodiversity indices of mosquitoes in Kurdistan Province, western Iran.

Methods: This study was carried out in 10 counties of Kurdistan Province. The immature stages of mosquitoes were collected monthly from June to September. ArcGIS software was used to spatial analysis and create maps. Alpha diversity indices were calculated using the related formula.

Results: Totally, 5831 larvae belonging to the family Culicidae were collected. Twelve species were identified including: *Anopheles claviger*, *An. maculipennis* s.l, *An. superpictus* s.l, *Culiseta. longiareolata*, *Cs. subochrea*, *Culex hortensis*, *Cx. mimeticus*, *Cx. perexiguus*, *Cx. pipiens*, *Cx. theileri*, *Cx. modestus* and *Cx. territans*. Based on this analysis, the high-risk areas of the province are determined as *Anopheles* in the west, *Culex* in the north, and the *Culiseta* in the south of the province. Analyzing the Alpha biodiversity indices showed Baneh and Sarabad had the maximum and Bijar had the minimum mosquito biodiversity.

Conclusion: The western counties of the province are regarded as the hotspots for anopheline mosquitos. Moreover, reporting of malaria cases in the past, bordering with Iraq and the high traffic of travelers have made these areas as potential foci for malaria transmission. So that, routine entomological inspections are proposed to detect any suspicious vector or case entrance.

Keywords: Mosquitoes; GIS; Diversity; Diptera; Larvae

Introduction

Mosquitoes (Diptera: Culicidae) have always been the focus of entomological studies due to their important role in transmitting a wide range of viral and parasitic diseases to humans or animals. More than half of the world's population lives in areas at risk of mosquito-borne diseases such as malaria, dengue fever, chikungunya, West Nile fever, Japanese encephalitis, and filariasis (1). Malaria is a parasitic infection transmitted by anopheline mosquitoes. It is estimated that 219 million new cases are reported worldwide, and lead to the death of more than 400,000 people annually. Dengue is the most common viral disease transmitted by *Ae-*

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des mosquitoes. More than 3.9 billion people in more than 129 countries are at risk of getting dengue fever, with an estimated 96 million symptomatic cases and an estimated 40,000 deaths each year (2). Malaria is also the most important mosquito-borne disease in Iran, which occurs mostly in the southern provinces of the country (3, 4). Seven species of *Anopheles* are among the most important vectors in Iran, including: *Anopheles stephensi* Liston, 1901, *An. culicifacies* sensu lato (s.l.) Giles, 1901 (5), *An. maculipennis* s.l. Meigen, 1818 (6), *An. fluviatilis* s.l. James, 1902, *An. sacharovi* Favre, 1903, *An. dthali* Patton, 1905, and *An. superpictus* s.l. Grassi, 1899 (7, 8). The presence of West Nile and Sindbis viruses, which are transmitted by mosquitoes, has also been reported in the country (9–13). Recently, mosquito-borne *Dirofilaria* (*D. immitis* and *D. repens*) has been reported in Iran (14).

In recent years, no local transmission of malaria has been reported in Kurdistan Province, and only in 2020, one case was reported in the city of Marivan, which was Iraqi (unpublished data from Department of Disease Control, Deputy of Health, Kurdistan University of Medical Sciences). In 2013 serologically positive cases of dengue fever was reported from Kurdistan Province, who had not travelled abroad (15); although, there is no report of occurring the vector *Ae. albopictus* or *Ae. eagypti* in this province. Till now no human case of Rift Valley fever virus has been reported in the province, though one study showed infection in sheep (16). West Nile virus is a widespread mosquito-borne arbovirus in Iran as it is reported in horses from at least 26 of the 31 Iranian provinces including Kurdistan (9). Different blood-feeding arthropods including mosquitos are considered as suspected vectors of tularemia which caused by the bacterium *Francisella tularensis*. Natural infection of tularemia in the wild reservoir, rodents (17), and in human have been reported in Kurdistan Province (18). The latest updated checklist of Iranian mosquitoes includes 70 species that represent eight or 12 genera, depending on the general classification of aedines (19, 20). Zaim (21) mentioned two genera and eight species of the subfamily Culicinae in Kurdistan Province. In previous studies conducted in the province, 18 species of the family belonging to five genera have been reported, which include *An. maculipennis s.l.*, *An. superpictus s.l.*, *An. claviger*, *An. sacharovi*, *An. algeriensis*, *An. marteri*, *An. sergentii*, *Coquillettidia richiardii*, *Culex hortensis*, *Cx. mimeticus*, *Cx. pipiens*, *Cx. theileri*, *Cx. perexiguus*, *Cx. territans*, *Culiseta longiareolata*, *Cs. subochrea*, *Ae. vexans* and *Ae. caspius* (22–24). In recent years, due to the reporting of dengue vectors in the south of the country (25, 26), and on the other hand, high reporting cases of dengue fever in eastern neighbors such as Pakistan, the importance of routine entomological studies has doubled in other border provinces to monitor the entrance of dengue vector/s. Sharing border with Iraq, made Kurdistan Province as one of the most important import destinations for goods from southeast Asian countries. One way of entering the dengue fever vector/s into the countries is through goods such as bamboo plant and car tires, therefore, conducting entomological studies in this critical point seem necessary. The studies that have been done in the province so far are only faunistic and scattered. The purpose of this study was to conduct a comprehensive study and update the species composition of mosquitoes in all counties of the province; moreover, to analyze the spatial distribution and to determine biodiversity indices of the mosquitoes throughout the province.

Materials and Methods

Study area

This study was conducted in 10 counties of Kurdistan Province in summer 2019. It is in the west of Iran between 34°44′N and 36°28′N and 45°33′E and 48°15′E. The area is 29.137 square kilometers, equivalent to 1/7 of the total area of Iran. This province, which is

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located in the scattered slopes and plains of the Middle Zagros Mountains, is limited from the north to the provinces of West Azerbaijan and Zanjan, from the east to Hamedan and Zanjan, from the south to Kermanshah, and from the west to Iraq. Kurdistan Province has 10 counties include: Sanandaj, Saqez, Marivan, Baneh, Qorveh, Kamyaran, Bijar, Divandarreh, Dehgolan and Sarvabad (Fig. 1). According to the general population and housing census of 2016, Kurdistan Province has a population of 1,603, 011 (Statistical Center of Iran). The highest and lowest average air temperatures in 2019 are belong to Sanandaj with 14.8 °C and Zarrineh with 8.8 °C, which they have different height from sea level. Marivan with 913.1mm and Qorveh with 352.7mm had the highest and lowest rainfall, respectively (Statistical Center of Iran).

Collection, mounting and identification of mosquito larvae

The immature stages of mosquitoes were collected monthly, for at least four months (once a month) from June to September. Larval collection was conducted in an urban point and four rural points at a distance of 15–20km in all counties. In fixed urban and rural points, three fixed and three variable locations were sampled. The distribution of sampling points included urban, rural, cultivated areas, and riversides. The mean water temperature and ambient humidity were recorded during the sampling period. The larvae of the mosquitoes were collected by standard dipping technique. Collected larvae were stored in lactophenol medium, and after clarification of morphological characters, they were mounted in de Faure's medium. After that, the larvae were identified using valid morphological Iranian identification keys (27). Additionally, the geographical coordinates of the sampling locations were recorded using a GPS device.

Collecting larvae using Ovitraps

Ovitrap is one of the most common techniques for collecting eggs and larvae of *Aedes* mosquitoes. In this study buckets, which were designed in a standard way, were used. Dark buckets, preferably black due to its attractiveness for *Aedes* mosquitoes, in a volume of two liters were used. After pouring two liters of water inside the buckets, 3 wooden pedals with dimensions of 15×5cm and a thickness of one to two mm were immersed in the water, and then they were attached to the wall of the bucket with clamps (28). All the information in the labels of the traps and the exact addresses of their installation places were recorded in a separate booklet. The traps were checked twice a week according to the weather conditions of the study areas. Due to evaporation of water inside the traps, water was added to them if necessary. During twice-a week check-up, the pedals were carefully examined for the presence of eggs by a handy lens. If a suspicious case of eggs was observed, the pedals were carefully removed and transported to the laboratory, and it was replaced by a new pedal.

GIS analysis

ArcGIS 10.4.1 software (http://www.esri.com/arcgis) was used to spatial analysis and creating maps. After inserting geographic coordinates into the Excel software, they were entered into the ArcMap in ArcGIS 10.4.1 software. Inverse Distance Weighted (IDW) interpolation analysis was employed to prepare raster maps.

Data process and analysis

To determine alpha diversity indices, data were inserted in Microsoft office Excel 2016 software using related indices, and coefficients showing below. Then, the average and standard deviation of all indices were calculated (29,30), and their P-value was determined by performing the analysis of variance (ANOVA) test in IBM SPSS Statistics 26 software.

Shannon–Weiner index: $H' = \sum_{i=1}^{S}(P_i)(\mathrm{Ln}P_i)$

H': species diversity index, S: number of species, pi: proportion of individuals of each species belonging to its species of the total number of individuals.

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Published Online: Dec 31, 2022

Evenness index: $E_{H'}=\frac{H'}{\log_3 S}$

H': species diversity index, S: number of species

Simpson's Index: $D = \frac{\sum n(n-1)}{N(N-1)}$

n: the total number of organisms of a particular species, N: the total number of organisms of all species

Menhinick's index: $D_{Mn} = \frac{S}{\sqrt{N}}$

S: number of species, N: the total number of organisms of all species

Margalef's index: $M = \frac{(S-1)}{lnN}$

S: number of species, N: the total number of organisms of all species

Hill: $N_1 = e^{H'}$

e: Napier's Constant, H': species diversity index

Hil2: $\frac{1}{D} = \frac{1}{\sum p_i^2}$

pi: proportion of individuals of each species belonging to its species of the total number of individuals.

Results

Species composition

During this study, a total of 5831 larvae belonging to the family Culicidae were collected from 56 localities of 10 counties of the province (Table 1–2). Of these, the highest number were from Marivan County and the lowest number were from Kamyaran County (Table 2). The collected larvae belonged to three genera: *Anopheles* 889 (15.24%), *Culiseta* 1826 (31.31%) and *Culex* 3116 (53.43). Collecting methods through ovitraps detected only a few larvae, which is belonging to the genus *Anopheles*, and no *Aedes* species were identified. 12 species of Culicidae were identified including: *An. claviger*, *An. maculipennis* s.l., *An. superpictus* s.l., *Cs. longiareolata*, *Cs. subochrea*, *Cx. hortensis*, *Cx. mimeticus*, *Cx. perexiguus*, *Cx. pipiens*, *Cx. theileri*, *Cx. modestus* and *Cx. territans* (Table 2).

Spatial analysis

Species belonging to *Anopheles* were identified in more than 50% of the counties with high abundance. The Baneh County, the western region bordering Iraq, has the highest number of species of this genus, about 2–3 times higher than other counties. Species of the genus *Culex* were caught in 60% of the counties. Divandere and Saqez have the highest abundance and are considered as high-risk areas for this genus. Species of *Culiseta* were identified in more than 50% of the study areas, which was abundant in the central parts of the province. In general, the abundance of *Culiseta* was higher than that of *Culex* and *Anopheles* (Fig. 2).

Inverse Distance Weighted (IDW) analysis

The results of IDW for *Anopheles* species in the study area showed that there was a very extensive hotspot in the west of Kurdistan Province. High density of *Anopheles* species in these areas increase the risk of malaria transmission in case of detecting vector infection. These high-risk areas locate in three counties bordering Iraq. Two small hotspots were determined in Saqez for the genus *Culex*, which is limited to the county itself, and is slightly extended to the north of the province.

On the one hand, the genus *Culiseta* has a medium hotspot, which exists in the south of the province and in the county of Sanandaj.

Based on this analysis, the high-risk areas of the province are determined as for *Anopheles* in the west, for *Culex* in the north, and for the *Culiseta* in the south of the province (Fig. 3).

Alpha diversity indices

Based on the calculation for the Simpson index, the maximum biodiversity was observed in Sarvabad and the minimum in Bijar. The maximum biodiversity, based on Shannon-Weiner index, was determined in Baneh and minimum in Bijar. Shannon-Weiner Evenness index showed maximum and minimum evenness of species distribution in Qorveh and Bijar, respectively. For Hill 1 and Hill 2 index the max-

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imum and minimum calculated in Baneh and Bijar respectively. The amount of species richness, based on Margalef's index, was maximum in Baneh and minimum in Bijar. Based on Menhinick's index, the maximum and minimum species richness was reported in Kamyaran and Bijar respectively. The statistical analyzes showed that there is a significant difference between the counties in all calculated indices (Table 3).

Table 1. Geographical coordinates of mosquito larval collecting sites, Kurdistan Province, summer, 2019

County	Location	Latitude	Longitude	County	Location	Latitude	Longitude
Sananadaj	Sanandaj	35.320776N	46.967762E	Bijar	Gharatoreh	35.8070749N	47.454365E
	Tavrivar	35.128055N	46.983173 E				
	Sarabghamish	35.329604N	47.047870E		Nadri	35.7606463N	47.529502E
	Kanimeshkan	35.237838N	46.921330 E		Sadeghabad	35.6553119N	47.169327E
	Babariz	35.364240N	47.065193E		Khorkhoreh	35.9689153N	47.800325E
	Doiseh	35.455993N	46.895943E		Salavatabad	35.9983444N	47.550961E
	Salavatabad	35.279508N	47.126181E		Ghabasorkh	35.8569962N	47.408851E
	Hasanabad	35.260319N	46.968247E		Mehrabad	35.9011767N	47.913803E
	Naran	35.154100N	47.067685E		Sayedan	35.9409092N	47.725527E
Baneh	Nojneh	36.128394N	45.783805E		Khosroabad	35.5182203N	47.623157E
	Aloot	36.025676N	45.571554E		Najafabad	35.7932236N	47.238271E
	Savan	36.074269N	45.904967E		Ghadimkhan	35.6248274N	47.642770E
	Ashtarabad	35.844582N	45.917856E		Ghamchaghai	36.1659343N	47.625592E
	Showe	36.044065N	45.865924E	Dehgolan	Bolbanabad	35.1404632N	47.321070E
Marivan	Vilae	35.592742N	46.307395E		Miraki	35.4350278N	47.290693E
	Chashniabad	35.653148N	46.031590E		Sis	35.2065655N	47.279559E
	Darahtephi	35.537565N	46.099041E		Bagahjan	35.3491741N	47.455589E
Sarvabad	Daranakhi	35.392268N	46.233966E	Qorveh	Mehdikhan	35.3456197N	47.649151E
	Ghalaji	35.358081N	46.282911E		Farhadabad	35.4154735N	47.616394E
	Rezab	35.260049N	46.403304E		Ghaleh	35.1335135N	47.803795E
Saqez	Pole-gheshlagh	36.091621N	46.340016E		Majin	35.1711562N	47.946798E
	Siyahdare	36.139224N	45.984306E		Bahraloo	35.195755N	48.121763E
	Kandalan	36.256530N	46.067004E		Sarab	35.1344474N	47.790924E
	Khanemiran	36.127169N	46.590750E	Kamyaran	Bovanah	34.8655297N	46.956129E
	Cheshme-Saqez	36.262425N	46.300410E		Shirvanah	34.7959729N	46.964364E
Divandarreh	Gavshalh	36.016072N	47.147437E		Kamyaran	34.7966668N	46.940417E
	Kolah	35.789951N	47.052898E		Alak	34.8033522N	46.843097E
	Hazarkaniain	35.769183N	46.813742E				
	Aghajari	35.891579N	47.128461E				
	Divandarreh	35.916419N	47.031293E				

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Table 2. Number of species belonging to mosquitoes in the counties of Kurdistan Province, summer, 2019

Species	County										Total (%)
	Sanandaj	Baneh	Marivan	Sarvabad	Kamyaran	Bijar	Saqez	Divandere	Qorveh	Dehgolan	
An. claviger	0	95	15	0	6	0	0	4	0	0	120 (2.05)
***An. maculipennis* s.l.**	3	136	269	52	1	0	0	52	0	0	513 (8.79)
***An. Superpictus* s.l.**	2	161	37	40	15	0	0	1	0	0	256 (4.39)
Cs. longiareolata,	885	57	125	75	86	217	58	0	1	313	1817 (31.16)
Cs. subochrea	1	0	8	0	0	0	0	0	0	0	9 (0.15)
Cx. hortensis	116	78	41	29	1	0	1	0	4	43	313 (5.36)
Cx.mimeticus	9	20	25	31	12	0	1	0	22	2	122 (2.09)
Cx.perexiguus	0	0	96	39	0	0	5	7	0	0	147 (2.59)
Cx.pipiens	20	56	80	13	29	0	440	96	27	8	769 (13.18)
Cx.theileri	7	18	573	124	16	1	103	416	153	27	1438 (24.66)
Cx. modestus	2	89	0	0	0	0	0	0	1	0	92 (1.57)
Cx.territans	0	28	0	0	0	0	40	167	0	0	235 (4.03)
Total	1045	738	1269	403	166	218	648	743	208	393	5831 (100)

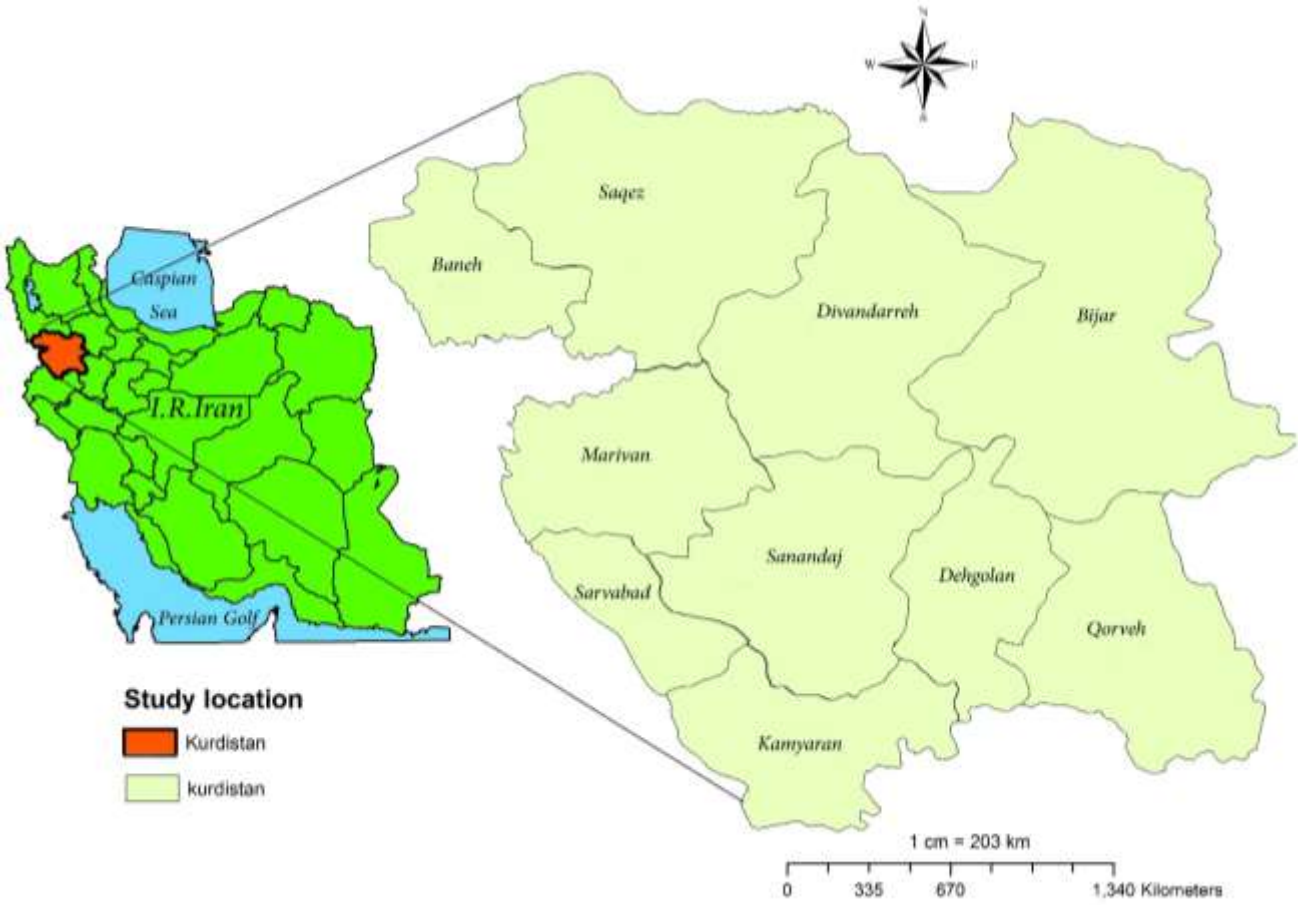

Fig. 1. Map of the mosquito larval collecting sites in Kurdistan Province, 2019

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Published Online: Dec 31, 2022

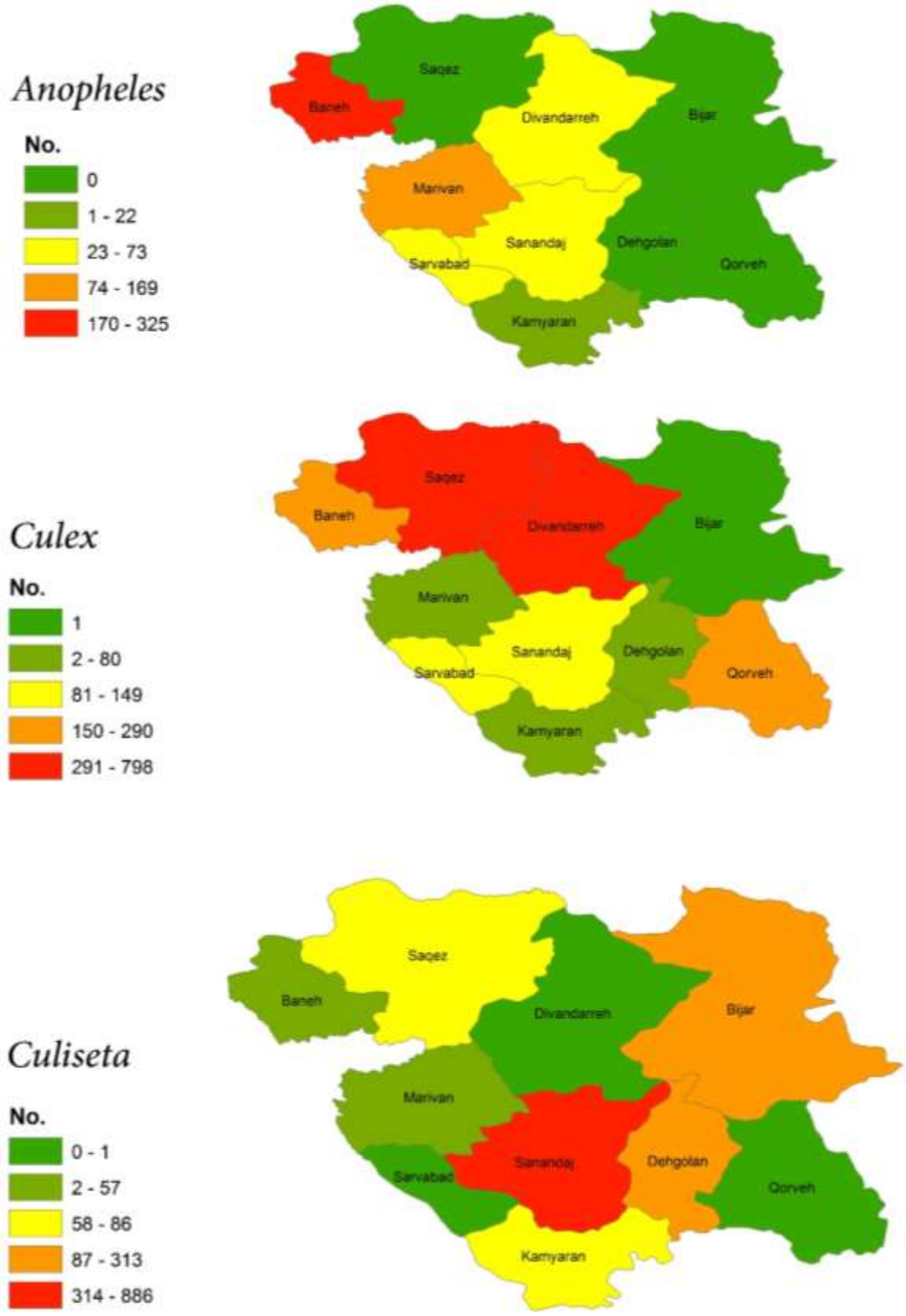

Fig. 2. Distribution of the mosquito genera belonging to Culicidae in counties of Kurdistan Province, summer, 2019

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Published Online: Dec 31, 2022

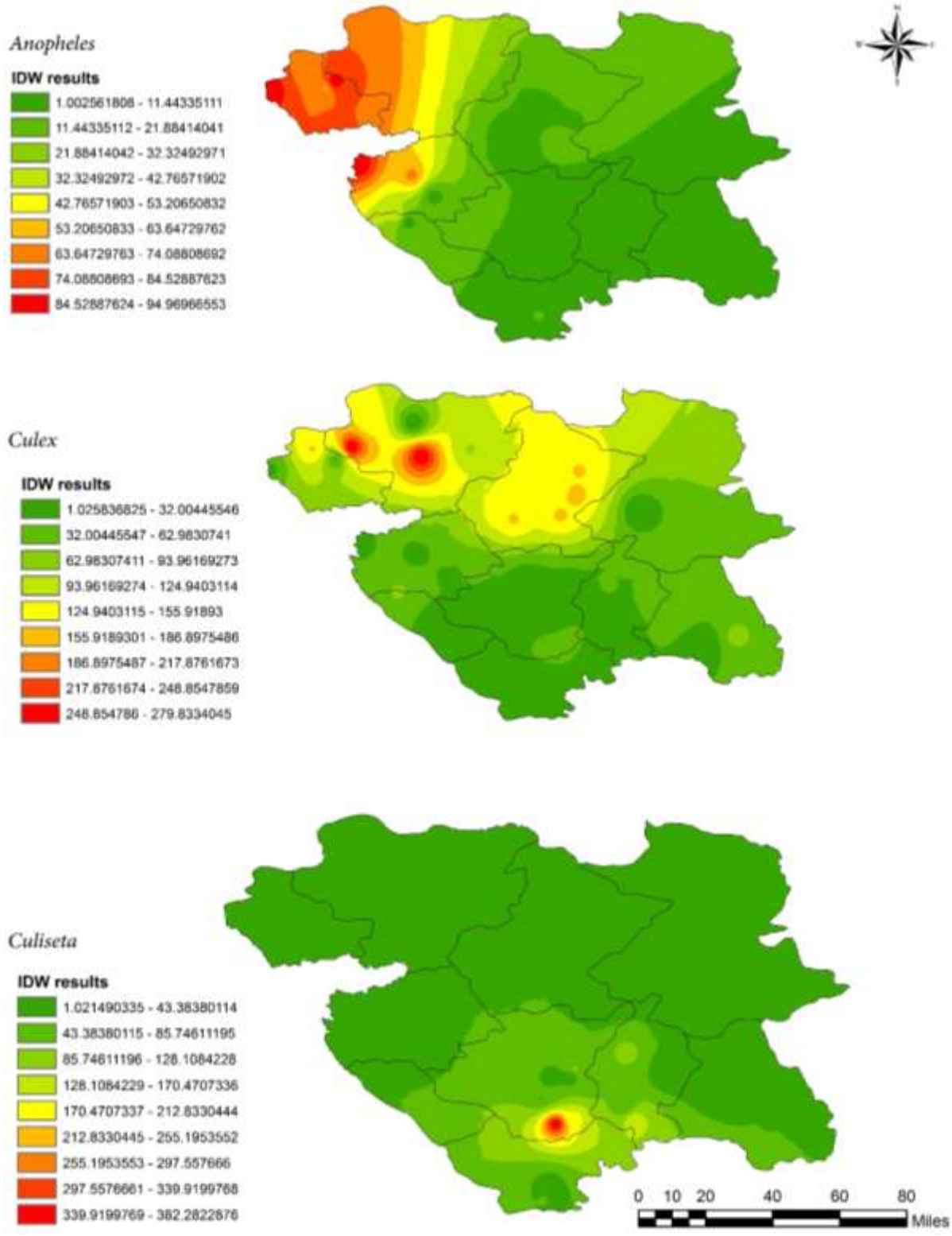

Fig. 3. Distribution of hot spots of the mosquito genera belonging to Culicidae in Kurdistan Province, summer, 2019

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Published Online: Dec 31, 2022

Table 3. The alpha diversity for mosquito species in Kurdistan Province, summer, 2019

County	Sympson Mean (SD)	P-Value*	Shanoon Mean (SD)	P-Value	Shaneven Mean (SD)	P-Value	Hil1 Mean (SD)	P-Value*	Hil2 Mean (SD)	P-Value	Margalef Mean (SD)	P-Value	Mnahenic Mean (SD)	P-Value
Bane	0.36 (0.36)		1.66 (0.44)		0.45 (0.26)		5.31 (2.84)		4.40 (2.37)		1.41 (0.81)		0.65 (0.31)	
Bijar	1.00 (0.02)		0.01 (0.04)		0.007 (0.026)		1.01 (0.04)		0.93 (0.26)		0.03 (1.00)		0.35 (0.09)	
Dehgolan	0.71 (0.23)		0.53 (0.39)		0.84 (0.59)		1.80 (0.61)		3.20 (3.72)		0.53 (0.36)		0.39 (0.09)	
Divandareh	0.50 (0.15)		0.96 (0.26)		1.14 (0.43)		2.68 (0.70)		2.13 (0.69)		0.90 (0.21)		0.43 (0.07)	
Ghorveh	0.67 (0.23)	P< 0.001	0.57 (0.41)	P< 0.001	1.16 (0.71)	P< 0.001	1.91 (0.84)	P< 0.001	1.63 (0.64)	P< 0.001	0.57 (0.37)	P< 0.001	0.58 (0.13)	P< 0.003
Kamyaran	0.48 (0.30)		1.01 (0.63)		0.44 (0.18)		3.18 (1.90)		2.66 (1.67)		1.09 (0.57)		0.82 (0.28)	
Marivan	0.32 (0.08)		1.41 (0.29)		0.49 (0.11)		4.22 (1.11)		3.26 (0.78)		1.25 (0.18)		0.54 (0.17)	
Saghez	0.61 (0.11)		0.69 (0.20)		0.34 (0.08)		2.03 (0.42)		1.68 (0.31)		0.68 (0.28)		0.40 (0.17)	
Sanandaj	0.76 (0.15)		0.46 (0.23)		0.26 (0.18)		1.62 (0.33)		1.35 (0.25)		0.64 (.41)		0.47 (0.23)	
Sarvabad	0.28 (0.06)		1.42 (0.28)		0.58 (0.08)		4.24 (1.07)		3.58 (0.80)		1.13 (0.40)		0.68 (0.23)	

*Probability from Kruskal-Wallis test

Discussion

A total of 5831 mosquitoe larvae were collected, which included 12 species and three genera. The highest number of species caught belonged to *Cs. longiareolata* (31%), and the lowest number belonged to *Cs. subochrea*. The highest number of *Anopheles* caught was from the border county of Baneh. This city has been one of the foci of malaria transmission (unpublished information, Kurdistan Province Health Center). Based on the maps that, generating using Arc GIS software, Baneh has been identified as one of the most important hot spots in terms of the existence of *Anopheles* species. Since, this county is an official border with Iraq, and on the other hand, is one of the well-known border markets in the whole country, it annually receives many travelers from inside and outside the country. Due to the presence of potential vector of the disease in this area, presence of a positive case of malaria may lead to a local transmission. Therefore, this issue requires a special attention to borders control and applying strict quarantine laws at the borders and entry points. On the other hand, routine entomological studies are required in this county, as well. In a recent study, the highest

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number of *Anopheles* specimens caught was reported from Sarvabad City (24). The highest number of *An. superpictus* s.l. specimens in our study was reported from Baneh City; while, in a recent study from Sarvabad City (24). *Anopheles superpictus* s.l. and *An. maculipennis* s.l. have been reported as the predominant species in Hamedan and West Azerbaijan Provinces, respectively (31, 32). Moreover, in East Azarbaijan Province, *An. maculipennis* s.l. has been reported as the predominant species of the genus *Anopheles* (33). Over the past few decades, *An. superpictus* s.l., *An. maculipennis* s.l. and *An. sacharovi* have been reported from 29, 20 and 18 out of 31 provinces of the country, respectively. *Anopheles superpictus* s.l. is more common in the western, and *An. maculipennis* s.l. and *An. superpictus* s.l. are more common in the north and northwest of the country (34).

In this study, more than 50% of the caught species belonged to the genus *Culex*. Based on IDW analysis, two high-risk points for this genus were identified in north-western areas of the province. The highest number (46%) belonged to *Cx. theileri*, and the lowest (2.9%) belonged to *Cx. modestus*. *Culex theileri* mostly caught from Marivan City in the west of the province. This species has recently been identified as a vector of *Dirofilaria* in the north of the country (35). In our study, seven species of the genus *Culex* were identified. In two other studies conducted in the province, four and five species of this genus were reported, respectively. In contrary to our study, *Cx. modestus* was not reported, and this species introduced for the first time in the province. In agreement to the result of our study, a study introduced *Cx. theileri* as the predominant species in the province (24). In another study, the highest number of caught species, in contrast to our study, was *Cx. theileri* (24, 36). In another study conducted in Sanandaj County, the predominant species caught in larval stage was *Cx. pipiens*, and in adult stage, like our study, was *Cx. theileri* (22). Like our study, in Hamedan, the predominant species was *Cx. theileri* (37), and in West Azerbaijan, *Cx. pipiens* was the predominant species (38). In East Azarbaijan Province, *Cx. theileri* species has been the most dominant species in the region (33). In the present study, two species of the genus *Culiseta* reported including *Cs. longiareolata* and *Cs. subochrea*. *Culiseta longiareolata* had the highest abundance (31.16%) among all species. This species was predominant in Sanandaj City, which previous studies in this city confirm the same results (22). In a study conducted by Banafshi et al. (12), in selected areas of Kurdistan Province, was reported as the dominant species. In Bijar, excluding *Cx. theileri* species, all the collected species were *Cs. longiareolata*, as the IDW analysis showed a hot spot for this species in this region. Similarly, *Cs. longiareolata* is the predominant species in Hamedan and West Azerbaijan Provinces (37, 38). In this study, no species belonging to the genus *Aedes* were reported by dipping technique or using ovitraps. Although, in previous studies a species belonging to this genus, *Ae. caspius*, had been caught in different parts of the province as well as neighboring provinces (22, 24, 33, 36, 38).

Calculation of biodiversity alpha indices showed that in most of the studied areas, the value of these indices is statistically different. This issue, in addition to the effect of confounding variables such as the manner and time of sampling, can be affected by different climates in the province. Different climates have created diverse ecological niches for the establishment of different species. The results of this study showed that areas with warmer climates had provided better conditions for higher biodiversity such as Sarvabad City. In studies conducted in the north of the country, Mazandaran province, and northwest, East Azerbaijan Province, similar to our study, different values were reported for biodiversity indices in the study areas (39, 40).

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Conclusion

To sum up, the western counties of the province are considered the hotspots for anopheline mosquitos. Moreover, reporting malaria cases in the past, bordering of these counties with Iraq, and the high traffic of travelers have made these areas as potential foci of malaria transmission. Although, in our study no species belonging to the genus *Aedes* were reported, previous studies have identified species belonging to this genus. Because two border counties of the province are the source of many goods importing from southeast Asian countries, which are endemic to dengue fever, routine entomological checks and rigorous quarantine inspections on entry points are necessary to detect any suspicious vector entrance.

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Ethical considerations

The study was approved by the Ethical Committee of Kurdistan University of Medical Sciences (IR.MUK.REC.1397/146).

Conflict of interest statement

The authors declare there is no conflict of interests.

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