

# Species Diversity and Larval Habitat Characteristics of Mosquitoes (Diptera: Culicidae) in Golestan Province, 2016

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## Abstract

**Background and objectives:** Mosquitoes (Diptera: Culicidae) are medically considered as one of the most important species of insects in terms of disease transmission to human beings; hence, they can be vectors of remarkable diseases such as malaria, West Nile, dengue fever, yellow fever, and filariasis. This study aimed to determine the species diversity and larval habitat characteristics of mosquitoes (Diptera: Culicidae) in Golestan Province in 2016.

**Methods:** In this descriptive cross-sectional study, larval habitats of mosquitoes were investigated in 14 counties of Golestan Province. Samples were collected by a standard ladle used for entomology. The collected larvae in lactophenol solution were transferred to a medical entomology laboratory in Gonbad-e Qabus County, and then microscopic slides were prepared. Mosquitoes species were identified based on their morphologic attributes and authentic identification keys. Larval habitat characteristics were studied in terms of the habitat type (permanent or temporary), water conditions (clear or turbid, stagnant or running), vegetation (with or without vegetation), sunlight exposure (full or partial exposure), and others. Data was analyzed using IBM SPSS software version 18.

**Results:** On the basis of analysis, 2891 third and fourth instars larvae, 4 genera (Anopheles, Culex, Culiseta, and Ochlerotatus), and 9 species (Anopheles superpictus, Culex mimeticus, Culex perexiguus, Culex pipiens, Culex pusillus, Culex theileri, Culex tritaeniorhynchus, Culiseta longiareolata, Ochlerotatus caspius) of mosquitoes were detected, with the Culex pipiens being recognized as the dominant species of this family in Golestan Province.

**Conclusion:** Due to the high species diversity of Culicidae in Golestan Province, further studies are of essence to investigate the ecology of medically important species such as Culex pipiens, Culex tritaeniorhynchus, and Anopheles superpictus, which are the vectors of many diseases in Golestan province.

**Keywords:** Culicidae, Larval habitat, Species diversity, Ecology, Golestan

## Introduction

Culicidaemosquitoes (Culicidae: Diptera) are known as one of the largest and most remarkable insect families. According to the most recent classifications, Culicidae encompass two subfamilies, 11 tribes, 112 genera, and 3539 species (1). According to a recent report, the systematic status of Culicidae in Iran contains seven genera, 16 subgenera, 64 species, and three subspecies (2). Over the recent years, some species have been added to this list. For example, *Anopheles superpictus* Grassi (2) was introduced as two separate species (3). Furthermore, a new species of *Anopheles hyrcanus* (i.e., *An. Hyrcanus* sp<sup>IR</sup>) was detected in Iran (4). Species such as *Aedes albopictus* Skuse and *Orthopodomyia pulczalpis* Rondani were also identified and reported in Iran for the first time (5, 6).

Over the past years, some research studies have also been conducted in Iran on the fauna and distribution of various species of mosquitoes in Golestan province. According to such studies, 25 species of mosquitoes have been reported in this province, the most medically important examples of which are *Aedes vexans*, *An. superpictus*, *An. maculipennis*, *Cx. pipiens*, and *Celsius theileri* (12-7).

The most commonly reported mosquito-borne diseases are Malaria and arbovirus infections such as West Nile Virus, Dengue Fever, Yellow Fever, and Filariasis, some of which are previously reported in Golestan province. Malaria is one of the major vector-borne diseases in Iran, which is transmitted by *Anopheles*. Over the past few decades, the northern regions of Iran, including the Golestan province, has been one of the focal points of malaria (14). Eight species of

*Anopheles* have been detected as the disease vectors in Iran, of which two species of *Anopheles maculipennis* and *Anopheles superpictus* were noticed in previous studies in Golestan province (11, 12). West Nile virus is also one of the leading vector-borne viruses transmitted by the *Culex* species to birds, horses and humans (15). Relevantly, some serological human and horse infections have been reported from Golestan province (16). *Dirofilaria immitis* is also one of the considerable pathogens transmitted by mosquitoes, reported in Golestan Province (17). In the northwest regions of Iran, *Culex theileri* has been reported as the vector of this filler (18). Given the importance of mosquitoes in the disease transmission, several studies have been carried out in the northern regions of Iran, including Golestan province, on their biology and ecology. Sofizadeh *et al.* (2016, 2017) reported the larval habitat characteristics of mosquitoes and biological attributes of *Anopheles* in Kalaleh County (11 & 12t). Azari-Hamidian *et al.* (2005, 2007, 2011) investigated the fauna and larval habitat characteristics of mosquitoes in northern and northeastern provinces in Iran (19-22). Nikookar *et al.* (2015, 2016, 2017) also carried out studies on fauna, larval nest characteristics and other biological properties of mosquitoes in Mazandaran province (23-25). According to the results of the aforementioned studies, various species of mosquitoes have different biological and ecological needs for growth, evolution, and survival. Some of these species lay eggs in temporary waters, and others lay eggs in permanent waters, some lay eggs in stagnant waters and some others do this in running waters. Some of these species prefer larval habitats with vegetation while some others prefer vegetation-free larval habitats. Some species prefer saline water, and some others prefer fresh water. Some species prefer

habitats with high temperature and humidity, while others live in lower temperature and humidity. To sum, some other differences are also observed in the developmental stages of mosquitoes; however, the commonality is that all species need water and larval habitats with water to grow and evolve and that their growth would be interrupted without water.

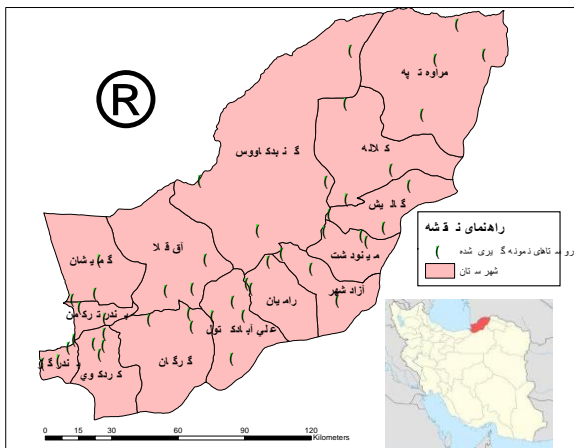
Considering the appropriate climate conditions in Golestan province for the development and growth of mosquitoes, and regarding the fact that there has been no comprehensive study in Golestan province on the biology and ecology of mosquitoes, the current study aimed to determine the species diversity and larval habitat characteristics of mosquitoes in Golestan province in 2016. The results would be implemented by the health authorities in this region as such they could control and prevent the mosquito-borne diseases.

## Materials and Methods

### *Setting of the study:*

Golestan province with an area of 204.47 square kilometers encompasses 1.3% of Iran's total area. The province (53° 51'-56 ° 21'4"E - 36° 30' 2" - 38° 7' 6"N) is located in the north of Iran. It is bordered to the north by the Republic of Turkmenistan, to the south by the eastern Alborz range and Semnan province, to the east by Northern Khorasan province, and to the west by the Caspian Sea and Mazandaran province. According to the last country divisions in 2010, the province is divided into 14 counties, 25 cities, 27 districts, 60 sub-districts (dehestans), and 1764 villages. Golestan province enjoys different climatic conditions because of its broadness and ecological diversity and its

being located within the eastern Alborz mountains from the south, Turkmenistan's warm and dry deserts from the north, the mountains of the northern Khorasan province from the east, and the Caspian Sea from the west. Regarding the existence of sea, forest, and mountains, Golestan province is climatically classified into temperate mountainous, cold mountainous (at altitudes above 3000 m), temperate Mediterranean, and warm semi-arid regions so that the farther we get from the south toward the north, the higher relative decrease in rainfall and humidity and the higher temperature increase we will notice, compared to the southern regions of the province. In terms of roughness, the province is divided into three distinct mountainous, foothill, and plain regions. The mountainous region is located in the south of the province and the highest mountains of the province are located in this area. Foothill regions in the southern and eastern parts of the province are located on the slopes and foothills of the highlands and fan- or cone-shaped deposit of small and coarse sediment cross and build up this region. Due to high soil permeability in the foothill regions, underground aquifers with abundant water are widely exploited as wells and aqueducts. Plain regions of the province are built up due to the withdrawal of Mazandaran Sea, severe erosion of the water, and the density of river alluvium. The lowlands are mostly located in this part of the province (with Caspian coastline 32 meters below the free water level). Population of this province mostly resides in the plains and mountainous regions (26).



**Figure 1.** Golestan province and sampled villages

### **Procedure:**

In this descriptive cross-sectional study carried out in Golestan province from May 2009 to September 2009, a minimum of three villages were selected in each county for sampling according to their topographic conditions and proximity to each other. Sampling in each village was performed in two steps: (1) Investigating larval habitats of mosquitoes, and (2) Sampling ovitraps installed in each of the concerned villages.

### **1. Investigating larval habitats of mosquitoes in the villages:**

After inspecting the larval habitats of mosquitoes in each village, appropriate habitats were selected for sampling. First, the following characteristics of the habitats were recorded in some prepared forms: Type of larval habitats (permanent or temporary, running or stagnant, natural or artificial), water condition (saline or fresh, clear or turbid), nest floor (clay, sand, cement), vegetation (out of water, on water, and under water vegetation or without

vegetation), and nest sunlight condition (sunny, shade). Sampling in these habitats was carried out once a month using 350 ml standard ladles. The standard dipping method was used 10 times in each village and all third and fourth instars larvae in the ladle were then collected.

### **2. Sampling ovitraps installed in each of the concerned villages:**

We used CDC Ovitrap for sampling. The ovitraps consisted of three-liter black plastic buckets, inside which three pieces of  $2 \times 10 \times 2$ -cm wood were fixed (27 and 28). The buckets were filled with water up to their  $\frac{3}{4}$  capacity, and then ten buckets were installed in each village. Once a week, each of the ovitraps was monitored and third and fourth instars larvae were taken out of them, if available.

For the sake of transparency, the caught samples were stored (canned) in vials containing Lactophenol for one week. Some characteristics (namely village name, trap date, trapper's name and habitat code) were recorded on the vials. These vials were transferred to a medical entomology laboratory in Gonbad-e Qabus Medical Center. To detect Culicidae, the third and fourth instars larvae were separated from the lactophenol medium and were mounted on Berlese medium. After three days, the specimens were identified using the larval identification keys (29). The data was imported to the IBM SPSS software version

18, and percentages and frequency were used to describe the data.

## Results

In this study, 2891 larvae were caught, according to whose morphological characteristics, 2348 and 543 larvae were caught from larval habitats and ovitraps, respectively (Table 1). The caught larvae were classified into 4 genera (Anopheles, Culex, Culiseta, and Ochlerotatus), and 9 species (Anopheles superpictus, Culex mimiticus, Culex perexiguus, Culex pipiens, Culex pusillus, Culex theileri, Culex tritaeniohynchus, Culiseta longiareolata, Ochlerotatus caspius).

Culex pipiens was recognized as the dominant species of this family in Golestan Province as it was caught from all of the counties in this province. In terms of frequency, Culex tritaeniohynchus was ranked second and was caught from most of the concerned counties (Table 2). In this study, more larvae were caught from permanent larval habitats than the temporary ones and from larval habitats with stagnant water than the ones with running water. Furthermore, more larvae were caught from larval habitats with sunlight and those with clay floor (Table 3). In this study, more larvae were trapped from some natural larval habitats such as river banks, lagoons, and pits, and from artificial larval nests and some artificial habitats such as ovitraps, boats, farm streams, sewages, and paddy fields (Table 4).

**Table 1.** Various species of mosquitoes caught from larval habitats and ovitraps in Golestan province, 2016

Species	larval habitats	ovitraps	Total
	N (%)	N (%)	N (%)
<i>Anopheles superpictus</i>	95 (4)	1 (0.2)	96 (3.3)
<i>Ochlerotatus caspius</i>	16 (0.7)	0 (0)	16 (0.5)
<i>Culex perexiguus</i>	4 (0.2)	0 (0)	4 (0.1)
<i>Culex pipiens</i>	1547 (65.9)	336 (61.9)	1883 (65.1)
<i>Culex pusillus</i>	27 (1.1)	16 (2.9)	43 (1.5)
<i>Culex tritaeniohynchus</i>	480 (20.4)	190 (35)	670 (23.2)
<i>Culex theileri</i>	25 (1.1)	0 (0)	25 (0.9)
<i>Culex mimiticus</i>	114 (4.9)	0 (0)	114 (3.9)
<i>Culiseta longiareolata</i>	40 (1.7)	0 (0)	40 (1.4)
Total	2348 (100)	543 (100)	2891 (100)

**Table 2.** Various species of mosquitoes caught from the counties in Golestan province, 2016

Species County	<i>Anopheles superpictus</i>	<i>Ochlerotatus caspius</i>	<i>Culex perexiguus</i>	<i>Culex pipiens</i>	<i>Culex pusillus</i>	<i>Culex tritaeniorhynchus</i>	<i>Culex theileri</i>	<i>Culex mimiticus</i>	<i>Culiseta longiareolata</i>	Total
Maraveh Tappeh	14	0	0	98	0	31	4	0	40	187
Kalaleh	31	0	2	1	0	15	1	3	0	53
Gonbad-e Qabus	0	16	0	220	30	456	0	0	0	722
Galikash	1	0	0	318	13	121	20	63	0	536
Minudasht	0	0	0	58	0	0	0	0	0	58
Azadshahr	0	0	0	173	0	0	0	40	0	213
Ramian	46	0	0	22	0	19	0	8	0	95
Aliabad Katoul	0	0	0	166	0	0	0	0	0	166
Gorgan	0	0	0	4	0	0	0	0	0	4
Kordkuy	0	0	2	242	0	18	0	0	0	262
Bandar-e Gaz	0	0	0	10	0	10	0	0	0	20
Bandar-e Torkaman	4	0	0	505	0	0	0	0	0	509
Gomishan	0	0	0	66	0	0	0	0	0	66
Total	96	16	4	1883	43	670	25	114	40	2891

**Table 3.** Various species of mosquitoes caught from different larval habitats and characteristics of each habitat in Golestan province, 2016

Characteristics of habitats	Species									Total
	<i>Anopheles superpictus</i>	<i>Ochlerotatus caspius</i>	<i>Culex perexiguus</i>	<i>Culex pipiens</i>	<i>Culex pusillus</i>	<i>Culex tritaeniolynchus</i>	<i>Culex theileri</i>	<i>Culex mimiticus</i>	<i>Culiseta longiareolata</i>	
<b>Characteristics of habitat</b>										
Permanent	14	4	0	311	0	136	15	48	34	562
Temporary	81	12	4	1236	27	344	10	66	6	1791
Running water	0	4	0	236	0	34	15	95	0	384
Stagnant water	95	12	4	1311	27	446	10	19	40	1964
<b>Vegetation</b>										
Out of water	13	12	4	409	18	150	5	43	0	654
On water	2	0	0	210	8	193	2	2	0	417
Under water	44	4	0	24	0	4	0	0	0	76
Vegetation-free	36	0	0	904	1	133	18	69	40	1201
<b>Sunlight</b>										
Sunny	82	12	0	966	10	275	22	87	0	1454
Shady	0	0	0	153	3	73	0	0	6	235
Semi-sunny	13	0	4	182	14	40	2	9	34	298
Sunny-shady	0	4	0	246	0	92	1	18	0	361
<b>Floor</b>										
Clay	73	16	4	645	26	379	10	35	0	1188
Sand	22	0	0	93	0	13	15	79	0	222
Rock	0	0	0	0	0	0	0	0	34	34
Cement	0	0	0	268	0	0	0	0	6	274
Wooden boat	0	0	0	175	0	0	0	0	0	175
Fiberglass boat	0	0	0	104	1	72	0	0	0	177
Unused tires	0	0	0	18	0	0	0	0	0	18
Others	0	0	0	244	0	16	0	0	0	260
<b>Water</b>										
Clear	81	16	4	776	27	443	14	104	40	1505
Turbid	12	0	0	193	0	34	0	0	0	239

Muddy	2	0	0	578	0	3	11	10	0	604
Fresh	83	12	4	1303	27	435	21	114	40	2039
Saline	12	4	0	29	0	34	4	0	0	83
Brackish	0	0	0	215	0	11	0	0	0	226
<b>Type of habitat</b>										
Natural	37	4	0	490	14	99	23	105	34	806
Artificial	58	12	4	1057	13	381	2	9	6	1542
Total	95	16	4	1547	27	480	25	114	40	2348

**Table 4.** Various species of mosquitoes caught from natural and artificial habitats in Golestan province, 2016

Species		<i>Anopheles superpictus</i>	<i>Ochlerotatus caspius</i>	<i>Culex perexiguus</i>	<i>Culex pipiens</i>	<i>Culex pusillus</i>	<i>Culex tritaeniorhynchus</i>	<i>Culex theileri</i>	<i>Culex mimiticus</i>	<i>Culiseta longiareolata</i>	Total
Type of habitat											
<b>Natural</b>	Riverside	37	0	0	82	0	37	4	48	0	<b>208</b>
	River bed	0	4	0	6	0	18	11	55	34	<b>128</b>
	Swamp	0	0	0	12	0	0	0	0	0	<b>12</b>
	Meadow	0	0	0	0	0	23	1	2	0	<b>26</b>
	Creek	0	0	0	93	0	17	0	0	0	<b>110</b>
	Pit	0	0	0	133	0	0	0	0	0	<b>133</b>
	Marsh	0	0	0	164	14	4	7	0	0	<b>189</b>
<b>Artificial</b>	Besides the paddy fields	49	0	0	58	2	11	0	6	0	<b>126</b>
	Within the paddy fields	0	0	2	5	0	12	1	0	0	<b>20</b>
	Stream adjacent to paddy fields	9	0	2	46	0	22	1	3	0	<b>83</b>
	Sewage	0	0	0	160	0	0	0	0	0	<b>160</b>
	Others Farm streams	0	12	0	38	10	210	0	0	0	<b>270</b>
	Animal footprint	0	0	0	0	0	8	0	0	0	<b>8</b>
	Pool	0	0	0	45	0	0	0	0	0	<b>45</b>
	Farm Pool	0	0	0	19	0	3	0	0	0	<b>22</b>
	Dishes	0	0	0	56	0	0	0	0	0	<b>56</b>



Tires	0	0	0	20	0	0	0	0	0	0	<b>20</b>
Wooden boat	0	0	0	330	0	0	0	0	0	0	<b>330</b>
Fiberglass boat	0	0	0	159	1	115	0	0	0	0	<b>275</b>
Wastewater	0	0	0	85	0	0	0	0	0	6	<b>91</b>
Water leakage	0	0	0	6	0	0	0	0	0	0	<b>6</b>
Stream	0	0	0	30	0	0	0	0	0	0	<b>30</b>
Ovitrap	1	0	0	336	16	190	0	0	0	0	<b>543</b>
<b>Total</b>	<b>96</b>	<b>16</b>	<b>4</b>	<b>1883</b>	<b>43</b>	<b>670</b>	<b>25</b>	<b>114</b>	<b>40</b>	<b>40</b>	<b>2891</b>

## Discussion

In this study, nine species of mosquitoes were caught, all of which had been previously studied in Golestan province. Since the previous studies have reported 25 species of mosquitoes (7-12), it could be concluded that the diversity of species in the present study was low. This might be due to the fact that the study sampling was limited to inside and outside the villages. More species diversity would have been observed if the studied species had been caught from different geographical regions such as forests and plains.

In this study, the most frequently observed species was *Culex pipiens*, which accounted for 65.1% of all species. The other studies in Golestan province have also reported this species as the dominant species, accounting for 77.3% and 27.6% of the caught species (30, 12). In two studies, *Anopheles hyrcanus* in Mazandaran province (23) and *Anopheles maculipennis* and *Culex theileri* in Ardebil province (18) were introduced as dominant species. *Culex pipiens* has been observed in almost all provinces of Iran (31). In a similar vein, this species was reported from all the counties of Golestan province in

the present study. The larval habitats of this species were diverse in this study while they were mostly caught in artificial, temporary and temporary larval habitats such as sewages, marshes, and pits. The same findings were reported in another study conducted in Golestan province (30). In Kazemi *et al.*'s (2000) study in Isfahan, this species was mostly found in rice farms and natural habitats (32). Zaim *et al.* (1987) claimed that the larval habitats of this species in Iran were artificial and stagnant freshwaters, including the irrigation canals, pits, and barrels filled with rainwater (33). A remarkable point in investigating the larval habitats of the *Culex pipiens* and its characteristics was that this species was caught from different larval nests with different ecological conditions and that it was the only species caught from the gathered water at the bottom of the wooden boat. Furthermore, *Culex pipiens* accounted for 58% of the species caught from water at the bottom of the fiberglass recreational boats. This species also comprised 62% of the total species caught from ovitraps. Similarly, *Culex pipiens* in another study in Golestan province accounted for 96.3% of the species caught from ovitraps (30). Hence it could be concluded that this species highly adapts to different types of larval habitats, which causes

the distribution and high abundance of this species in Iran and all regions in Iran (31). This species is of high medical importance since it carries several pathogens such as *Wuchereria bancrofti*, West Nile viruses, Japanese Encephalitis, St. Louis, Sindbis, Western Equine Encephalitis, Eastern Equine Encephalitis, Rift Valley Fever, and so on (31).

*Cx. Tritaeniorhynchus* was another species caught from a majority of the counties in this province. In terms of frequency, this species was ranked second, accounting for 23.2% of the total caught larvae, 20.4% of the total larvae separated from the larval nests, and 35% of the total larvae caught from the ovitraps. These values reported in another study (12.4%, 21.7% and 3.1%, respectively) in Golestan province were similar (30). In accordance with the results of this study, this species was ranked second in Guilan province as well (31). On the contrary, no larvae of this species was observed in Mazandaran, Zanzan, and Kohgiluyeh and Boyer-Ahmad provinces (35, 34, 23). In this study, this species was caught from different larval nests and its frequency in Golestan province, in line with the results of the other studies, was higher in temporary larval nests with stagnant water than the permanent larval nests with running water (30). The frequency of this species in larval nests with the clay floor and sunlight was more than that in shadow larval nests with sandy or cement floor. *Culex Tritaeniorhynchus* and *Culex pipiens* were species caught in all the three habitats with fresh, brackish, and saline water. In Iran, this species has been observed in 13 provinces; however, its distribution was higher in Khorasan province, the Caspian Sea, East Azarbaijan province, and all southern regions of Iran (31). Medically, *Culex Tritaeniorhynchus* is of paramount

importance and carries several pathogens such as *Wuchereria bancrofti*, *Dirofilaria immitis* and Chikungunya viruses, Sindbis, West Nile, Japanese Encephalitis, and Dangué fever (31).

In the current study, *Cx. Perexigus* was caught in low frequencies from Kalaleh County in the eastern part of Golestan province and Kordkuy in the western part of this province. Other studies have pointed out that the species prefer temporary habitats to the permanent ones (36, 33). Similar results were obtained in the present study, and this species was only caught from the inner banks of paddy fields and their adjacent streams. In the same line, the preferred nest of this species in another study in Golestan province was found to be the inner banks of paddy fields (30).

*Cx. Mimiticus* is distributed in southern Palearctic and Oriental regions as well as in 16 provinces in Iran (31). Consistent with the results of another study in Golestan province, this species in the present study was only caught from the eastern counties of the province (Kalaleh, Galikash, Azadshahr, and Ramian) (30). In the present study, this species was caught from rivers with sandy beds; however, they in another study in Golestan province were found in marshes and water leakages with muddy beds (30). This species has not been introduced as a vector of certain diseases in medicine and health fields (37).

In the current study, *Cx. theileri*, known as a nationally-distributed species in Iran (31), was caught in a low frequency from the eastern counties of Golestan province (Maraveh, Kalaleh, and Galikash). Similar to the results of this study, this species was in low frequency in Gilan province (31); however, it was the dominant species in Ardebil (18), Zanzan (38), Hamedan, and

Kurdistan (39, 36). The preferred habitat for this species and *Cx. Perexigus* were similar as such the former was caught from rivers, marshes, meadows and paddy fields. This species was also caught from the larval nests with running water and nests with stagnant water, confirming the results of another study in Golestan province (30). In a study in Hamadan province, larval habitats of this species included permanent and stagnant marshes with inside, outside, and immersed vegetation (36).

In this study, *Cx. Pusillus* was caught from temporary and permanent larval nests such as marshes, farm streams, and water accumulated at the bottom of fiberglass recreational boats. In addition, this species along with *Culex pipiens*, *Culex Tritaeniorhynchus*, and *Anopheles Superpictus* were caught from the ovitraps; therefore, it could be concluded that this species also has acceptable adaptation to different larval nests.

In this study, we caught and identified the *An. Superpictus*; however, previous studies in Golestan province have reported the presence of other species, including *Anopheles maculipennis*, *Anopheles hyrcanus*, *Anopheles claviger*, *Anopheles plumbeus*, *An. pseudopictus*, *An. pulcherrimus*, *An. turkhudi*, *An. Multicolor*, and *An. algeriensis* (30, 12 and 11). In the present study, *An. superpictus* was collected from natural and artificial habitats such as riversides and banks of paddy fields with rocky and muddy beds. Consistent with the results of our study, this species in another study in Golestan province was collected from the riversides and riverbeds and rivers of permanent stagnant water with or without vegetation (30). The

same findings were reported in studies in Hamadan and Kalaleh (12, 36). Kazemi *et al.* (2000) in Isfahan stated the characteristics of the larval nests for *An. Superpictus* as deep and clear vegetation-free water with rocky beds as well as rivers with sandy beds and sunlight, and shallow streams with muddy beds (32). In the current study, the larvae of this species were also caught from both of these larval nests. Azari Hamidian *et al.* (2009) in their study in Ardabil reported a greater number of this species from permanent and temporary larval nests with immersing vegetation and from river bed holes (18); however, contrary to the results of their study, the species in the present study were not found in larval nests with immersing vegetation.

In this study, *Oc. caspius* was the only detected species of *Ochlerotatus*, which was found in Gonbad-e Qabus. This species has been found in different studies in Iran (18, 39) as it was reported in another study conducted in Golestan province (30). In Kalaleh, three species of this genus (namely *Oc. caspius*, *Oc. Echinus*, and *Oc. geniculatus*) were observed (12). Contrary to the results of this study in which this species was found to be in low frequency, this species in Kurdistan was the second most frequent larvae, following the *Culex theileri*. In the present study, we caught this species from rivers and farm streams. This species is medically significant and is known to be the vector of *Tahyna* and *Tularemia* and the potential vector of the Rift Valley fever virus and *Dirofilaria Immitis* (31).

Further, *Cs. Longiareolata* was the only species from the genus *Culiseta* found in Maraveh Tappeh. Little information is

available about the ecology of the *Culiseta* Iran. In this study, we caught the *Cs. longiareolata* from vegetation-free rivers and sewages with cement and rocky beds, as this species in a previous study in Golestan province was found in Kalaleh and a vegetation-free water storage with cement floor (30). This species, however, was reported from various larval nests in studies conducted in Iran in 1986 and 2007(2, 33). In Yazd county, Dehghan *et al.* (2011) also observed *Cs. longiareolata* along with *Culex pipiens* in larval habitats infested with organic materials and waste from factories, plants, and cement ponds used to store water for animals and livestock (40).

### Conclusion

Given the appropriate climate conditions in Golestan province, numerous larval habitats, and development of farming activities, various species of Culicidae mosquitoes grow in this province. Because some of them are the vectors of various diseases, the preconditions exist for the transmission of some diseases. In this study, we caught no definitive vector for diseases such as Dengue fever, Zika, and chikungunya. Further studies are recommended to find these vectors in Golestan province.

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### Declarations

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#### *Conflict of interest*

None

#### *Authors' contributions*

All authors contributed equally

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