

## Bionomics and Breeding Places of the Genus *Anopheles* (Diptera: Culicidae) in Mahroo and Sepid-Dasht Districts, Luristan Province, Western Iran

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Article information	Abstract
<p>Article history: Received: 5 Oct 2011 Accepted: 26 Apr 2012 Available online: 15 July 2012</p> <p>Keywords: Bionomics Breeding Place <i>Anopheles</i> Fauna Seasonal Activity Luristan Province Iran</p> <p>*Corresponding author at: Department of Medical Entomology and Vector Control, School of Health, Ahwaz Jundishapur University of Medical Sciences, Ahwaz, Iran. E-mail: <a href="mailto:Hamid.kassiri@yahoo.com">Hamid.kassiri@yahoo.com</a></p>	<p><b>Background:</b> Study on ecology and larval habitats of anopheline mosquitoes are important in terms of some aspects such as, vector control through manipulation and modification of these habitats that can reduce the burdens of mosquito-borne diseases. Given the likelihood of malaria epidemic, this work has been conducted to study anopheline mosquito fauna, larval habitat features and seasonal activity of the genus <i>Anopheles</i> in the Mahroo and Sepid – Dasht rural districts, Luristan province.</p> <p><b>Materials and Methods:</b> This descriptive cross-sectional investigation was done from April to November 2001-2002. Larvae were collected by means of dipper and dropper from breeding places using the standard dipping technique, every two weeks. The third and fourth instar anopheline larvae were preserved in Lactophenol and identified into species using morphological characters. Larval habitat characteristics were recorded.</p> <p><b>Results:</b> A total of 4376 <i>Anopheles</i> larvae were collected. Five species and two morphological forms were found which include <i>Anopheles superpictus</i> Grassi form A (76.3%), <i>A. dhali</i> Patton (9.7%), <i>A.turkhudi</i> Liston (8.5%), <i>A. marteri</i> Senevet and Prunelle (4%), <i>A. superpictus</i> Grassi form B (1.1%) and <i>A. apoci</i> Marsh (0.4%). In these districts, anopheline larval activity began in early May and ended in early November. In this paper, the characteristics of larval habitats of any species have been discussed separately.</p> <p><b>Conclusion:</b> The main larval habitats of important vector of the region, <i>A. superpictus</i>, were determined river sides, stream margins and rice fields. The findings of this research can be used to manage control of vectors.</p> <p>Copyright © 2012 Zahedan University of Medical Sciences. All rights reserved.</p>

### Introduction

Malaria is a protozoan parasitic disease that is caused due to *Plasmodium* and is transmitted by female *Anopheles* mosquitoes [1]. Over half the world's population in nearly 100 countries are at risk of malaria. It is estimated that annually about 300-500 million people are infected with this disease that 2-3 million of them, that are mostly pregnant women and children, will die [2]. Having been located in the Eastern Mediterranean region, Iran is one of malarious countries in the world where malaria is endemic [3]. About 60% of the population in this region are at risk of malaria where an average 10 million cases of malaria and 50 thousand deaths are annually reported. Almost 45% of the population of the Eastern Mediterranean region are living in the areas with transmission of *Plasmodium falciparum* and *P.vivax*, and 15% are living in areas with transmission of *P. vivax* [4, 5].

Most malaria cases in Iran are reported from South East of country and some cases are reported from border line regions between Iran and Azerbaijan in provinces of Guilan, Ardebil, East Azerbaijan and West Azerbaijan. In recent years, 11- 60 thousand cases of malaria are annually reported [6-8]. The cause of disease in Iran are *P. falciparum* and *P. vivax* species which are

approximately equal in terms of frequency, but in recent years *P. vivax* infection has a higher frequency than *P. falciparum* [9]. Resistance of vectors to insecticides, resistance of malaria parasite to drugs and administrative and managerial problems, still remain this disease as a serious problem in the world. Nowadays, control of malaria vectors is considered as one of the most important strategies to combat malaria. The basic strategy of the World Health Organization in this regard is the use of mosquito-nets impregnated with pyrethroid insecticides. This method could effectively reduce malaria morbidity and mortality by between 20-63% [10].

Study of biology, ecology and species composition of anopheline mosquitoes in each region is very necessary for malaria control planning. The genus of *Anopheles* includes four subgenera and at least 458 identified species in the world [11, 12]. According to the studies so far conducted in Iran, at least 26 species of genus *Anopheles* have been reported from two subgenera of *Anopheles* and *Cellia* [13]. Shahgudian and Lotfi provided the key to identify *Anopheles* and *Culex* mosquitoes in Iran [14, 15]. Dow has noted some ecological and biological characteristics of mosquitoes in Iran [16]. Zaim conducted extensive researches on mosquitoes in Iran [17, 18]. Azari

et al. studied characteristics of breeding places of mosquitoes in the county of Rasht [19]. Salehi et al. determined malaria vector *Anopheles* species and some of their ecological characteristics in the Farsan county [20]. Kayedi et al. published the results of examination of index of blood feeding from human for *Anopheles superpictus* collected from Luristan province [21]. Sedaghat and Harbach prepared a list of Iranian *Anopheles* and presented some information about their ecology [22]. Despite the conducted studies, there is little information about the bioecology of the different *Anopheles* species; especially *Anopheles superpictus*. Thus, the present investigation was conducted to study the ecology of *Anopheles* in the two districts of Mahroo and Sepid-Dasht respectively from suburbs of Counties of Aligudarz and Khorram-Abad.

### Materials and Methods

This was a descriptive cross-sectional study conducted to obtain information about *Anopheles* mosquitoes' fauna, distribution and characteristics of *Anopheles* larval habitats and seasonal activity of *Anopheles* larvae in counties Aligudarz and Khorram-Abad from April to November 2001-2002.

Luristan province, with an area of about 28157 square kilometers, is located in West of Iran between 46 degrees and 50 minutes to 50 degrees and 1 minute east longitude and 32 degrees and 40 minutes to 34 degrees and 23 minutes north latitude from the Greenwich meridian. Its average altitude is over 2200 meters above sea level. The lowest point of province with 239 m in height is located in plains of province and its highest peak, Oshtoran Kooch with an altitude of 4080 meters above sea level, is located among the Zagros Mountain. This province has 9 counties, 23 cities, 26 sectors and 83 districts. This province has three different climates including cold mountainous weather that has cold winters and mild summers, temperate region climate in which Khorram-Abad county is located and warm region climate that has warm summers and relatively favorable winters due to the effects of hot winds of Khuzistan province

Aligudarz county has two types of cold weather (in the central and northern area) and temperate and warm climate (in the southern part). The altitude of Khorram - Abad and Aligudarz is 1200 meters and 2000 meters above sea level respectively. The average annual rainfall in Khorram-Abad is 570 mm and minimum and maximum annual rainfall is respectively about 234 mm and 770 mm. Average annual rainfall in Aligudarz fluctuate between 450-800 mm. Khorram-Abad is located in 21 minutes and 48 degrees longitude and 29 minutes and 33 degrees latitude. Geographical position of Aligudarz is between 49 degrees and 42 minutes east longitude and 33 degrees and 24 minutes north latitude.

In this research, dipper and dropper were used to catch *Anopheles* larvae. In order to study fauna and the properties of breeding places of anopheline mosquitoes in the districts of Mahroo and Sepid-Dasht from April to late November, 51 numbers were collected *Anopheles* larvae from 5 villages in these districts (including Gooshe Chal

Bozorg, Margsar, Klondi, Keshvar and Tang Haft). Studies were conducted on seasonal activity of *Anopheles* larvae in the three villages of Gooshe Chal Bozorg and Margsar (Aligudarz County) and Klondi (Khorram-Abad County) once every 15 days and abundance of *Anopheles* larvae was determined in the larval nests by the number of larvae of 3 and 4 instars in 10 dippers. Larvae of each type of larval nests associated with each village after being washed with distilled water, were preserved separately in test tubes containing lactophenol. Full details including the type of larval nests, name of village and district, date of collection and name of the collector were recorded on test tubes. Larvae were kept in Lactophenol solution at least 2 days to become clear. Larvae were prepared in liquid Iodophor solution on slides. Samples were diagnosed with help of microscope using a valid identification key [14].

### Results

In the study of fauna determination, total of 4376 *Anopheles* larvae were caught in 51 times from 5 hamlets in districts of Sepid-Dasht and Mahroo. In the present investigation, five species of *Anopheles* and two morphological forms including *Anopheles (Cellia) superpictus* Grassi (1899), *Anopheles (Cellia) dthali* Patton (1905), *Anopheles (Cellia) turkhudi* Liston (1901), *Anopheles (Cellia) apoci* Marsh (1933) and *Anopheles (Anopheles) marteri* Keshishian and Pringle (1954) were caught which were described in table 1 by relative abundance and catch region.

*Anopheles superpictus* with 3386 numbers (77.4%) out of all caught mosquitoes had the most abundant and *Anopheles apoci* with 15 numbers (0.4%) had the lowest abundance. In this study, two completely distinct larval forms of *Anopheles superpictus* are reported. Morphological forms A and B of this species were observed respectively with relative abundance of 76.3% and 1.1%. *Anopheles superpictus* larvae form B compared to form A had two rows of frontal hairs, a collar, two pairs of Palmate hairs in each abdominal segment and oval accessory tergal plates. These two forms of *Anopheles superpictus* and *Anopheles apoci* are reported for the first time from these regions. Specifications of larval nests of known species (Table 2) are as follows:

*Anopheles superpictus* (form A): from 4376 caught *Anopheles* larvae, total 3339 ones (76.3%) were *Anopheles superpictus* (form A) which were found in all studied larval habitats (Table 2).

Among larval nests containing *Anopheles superpictus* larvae, 58.8% were permanent and 41.2% were temporary and 56.9% of larval nests with flowing water and 43.1% with stagnant water. *Anopheles superpictus* choosed its larval nests in sunny locations so that 82.3% of larval nests of this species were sunny and 62.8% had vegetation. About 62.7% of its larval nests were with a muddy bottom and the rest were with a sandy or rocky bottom. Approximately 94.1% of oviposition of this species occurred in clear waters.

The most important larval habitats of this species were respectively determined rivers edge (27.5%), streams

edge (23.5%) and plots of rice cultivations (21.5%). *Anopheles dthali*: 424 larvae of this species (9.7%) were collected. In permanent and current waters 59.4% and 68.7% were caught, respectively. This species laid its eggs in a variety of larval nests with or without plants, but it was often found in larval habitats with plants (71.9%). *Anopheles dthali* is a quite sun-loving species, so that over 87.5% of its larvae were collected from sunny breeding places. It is grown in the larval habitats whose bottom is often from mud (65.6%) and with clear water (96.9%). The breeding places of this species were mainly streams edge (28.1%) and rice cultivation plots (21.9%), respectively.

*Anopheles turkhudi*: This species which existed in 21 larval nests included a total of 8.5% of the collected larvae. *Anopheles turkhudi* was found more in permanent waters (61.9%) and current waters (71.4%) and 61.9% of its larval nests had plants. The above species selected habitats exposed to sunlight (90.5%) which had a mud substrate (61.9%) or sandy substrate (38.1%) and clear water (100%) for oviposition. Most larval nests of this species were streams (28.6%), along rivers (19%) and grasslands (19%), respectively.

*Anopheles marteri*: This species was found in 11 larval nests with relative abundance of 4%. It was found in permanent waters (54.5%) or temporary waters (45/5%) and mostly current waters (81.8%) with plant (54.6%) or without plant (45.4%). It was mainly collected from sunny larval nests (63.6%) and with clear water (90.9%). The dominant larval nests of this species were mostly along rivers (45.4%), streams (18.2%) and rice crop plots (18.2%), respectively.

*Anopheles superpictus* (form B): This species was observed in 24 larval nests with relative abundance of 1.1%. It was collected mostly from current waters (75%), exposed to sunlight (83.3%) and clear waters (91.7%). It was found in permanent larval nests (58.3%) or temporary (41.7%), with the plant (58.3%) or without plant (41.7%), mud bed (58.3%) or sandy bed (37.5%). *Anopheles superpictus* larval nests (form B), were often along rivers (33.3%), streams (29.2%) and rice crop plots (16.7%), respectively.

*Anopheles apoci*: Relative abundance of this species was 0.4% which was collected from 11 sunny larval nests (100%) with clear water (85.7%), often a mud bottom (71.4%) and plant eligible (57.1%) or no plant (42.9%). The larval nests of this species were determined permanent (57.1%) or temporary (42.9%) and with current water (42.9%) or stagnant water (57.1%).

*Anopheles apoci* was often caught from along rivers (42.9%) and rice crop plots (28.6%).

Larval activity of *Anopheles* species in Mahroo District began from the first half of May and ended in the first half of November. The maximum abundance of anopheline larvae was obtained in the first half of August until the second half of August, respectively, with the relative abundance of 101.2 and 101 the III and IV instars of anopheline larvae per 10 dippers (Fig. 1). Similarly, in the District Sepid-Dasht, seasonal activity of anopheline larvae began from the first half of May and ended in the first half of November. Anopheline larval activity peak in this district was in the first half of August and with the relative abundance of 115.2 the III and IV instars of anopheline larvae per 10 dippers (Fig. 2).

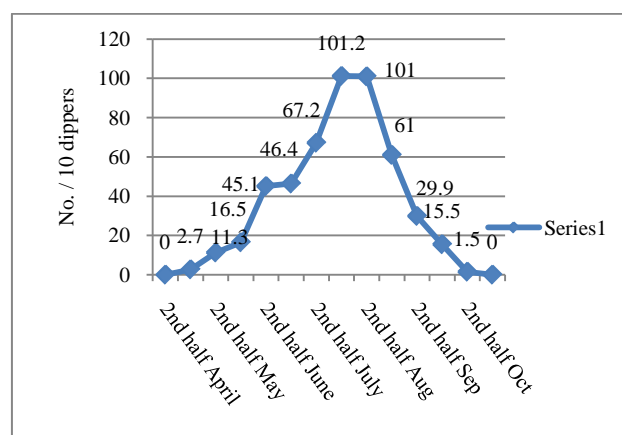


Figure 1. Monthly prevalence of anopheline larvae in Mahroo District, Aligudarz County, during 2001-2002

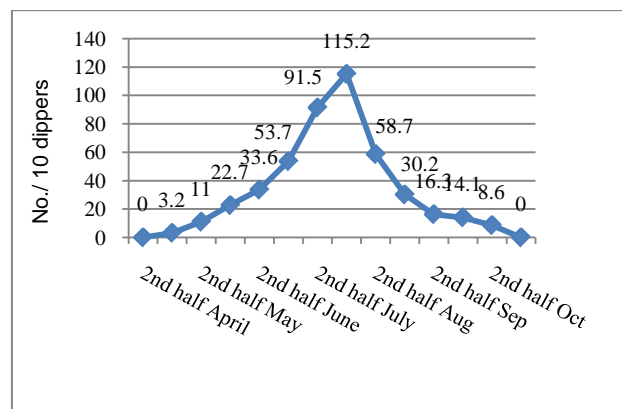


Figure 2. Monthly prevalence of anopheline larvae in Sepid-Dasht District, Khoram-Abad County, during 2001-2002

Table 1. Fauna and relative abundance of Anopheles mosquitoes in the Districts of Mahroo and Sepid-Dasht, Luristan province, Western Iran

Rural District Species	Mahroo N (%)	Sepid – Dasht N (%)	Total N (%)
<i>Anopheles superpictus</i> (form A)	2328(74.8)	1011(79.9)	3339(76.3)
<i>Anopheles dthali</i>	264(8.5)	160(12.6)	424(9.7)
<i>Anopheles turkhudi</i>	317(10.2)	57(4.5)	374(8.5)
<i>Anopheles marteri</i>	161(5.2)	16(1.3)	177(4)
<i>Anopheles superpictus</i> (form B)	33(1.1)	14(1.1)	47(1.1)
<i>Anopheles apoci</i>	8(0.2)	7(0.6)	15(0.4)
Total	3111(100)	1265(100)	4376(100)

**Table 2.** Ecological features of the breeding places and the occupation percentage of each one of them by *Anopheles* species in Districts of Mahroo and Sepid-Dasht, Luristan province, Western Iran

Anopheles species	Characteristics	Superpictus(form A)	Turkhodi	D'thali	Apoci	Marterii	Superpictus(form B)
Numbers of positive breeding places		51	21	32	7	11	24
Type of water	Permanent	58.8	61.9	59.4	57.1	54.5	58.3
	Temporary	41.2	38.1	40.6	42.9	45.5	41.7
Stream of water	Current	56.9	71.4	68.7	42.9	81.8	75
	Stagnant	43.1	28.6	31.3	57.1	18.2	25
Status of plant	Out of water	45.1	38.1	50	14.2	36.4	37.5
	The water level	11.8	19	15.6	28.6	9.1	16.7
	Under water	5.9	9.5	6.3	14.3	9.1	4.2
	Without plants	37.2	33.4	28.1	42.9	45.4	41.7
Sunlight status	Sunny	82.3	90.5	87.5	100	63.6	83.3
	Semi-Shade	11.8	0	6.3	0	27.3	12.5
	Shadow-Sunny	5.9	9.5	6.2	0	9.1	4.2
Bottom of breeding place	Muddy	62.7	61.9	65.6	71.4	45.5	58.3
	Sandy	31.4	38.1	28.1	0	36.4	37.5
	Rocky	5.9	0	6.3	28.6	9.1	4.2
	Turbid	5.9	0	3.1	14.3	90.9	8.3
Status of water	Clear	94.1	100	96.9	85.7	90.9	91.7
	Fresh	94.1	85.7	90.6	85.7	100	87.5
	Salty	5.9	14.3	9.4	14.3	0	12.5
	Spring	11.8	14.3	15.6	0	9.1	12.4
The natural larval habitats	Wetland	2	4.8	0	14.2	0	4.2
	Edge of river	27.4	19	15.6	42.9	45.4	33.3
	Grassland	11.8	19	15.6	0	9.1	4.2
	Edge of stream	23.5	28.6	28.1	14.3	18.2	29.2
	Plot of rice cultivation	21.5	9.5	21.9	28.6	18.2	16.7
Artificial larval habitat	Irrigation channel of farm	2	4.8	3.2	0	0	0

## Discussion

In the study of the fauna of anopheline mosquitoes in the Districts of Mahroo and Sepid-Dasht, respectively from suburbs of Aligudarz and Khorram-Abad, species of *Anopheles superpictus*, *Anopheles dthali*, *Anopheles turkhudi*, *Anopheles marteri* and *Anopheles apoci* were found among which *Anopheles superpictus* (77.4%), *Anopheles dthali* (9.7%) and *Anopheles turkhudi* (8.5%) had more relative abundance. In this research, species of *Anopheles apoci* and two morphological forms of *Anopheles superpictus* (A and B) are reported for the first time in this region which are quite different in terms of larval features and the differences concerning the presence or absence of the collar, the number of rows of frontal hairs, the number of Palmate hairs in each abdominal segment. *Anopheles superpictus* is one of the major malaria vectors in Iran which is widely spread in the country. In Iran, this species is caught as high as nearly 2,000 meters above sea level.

This *Anopheles* species has a wide distribution in Asia, Europe and North Africa. *Anopheles superpictus* is the important vector of Palearctic region, countries in North Africa, Middle East, India, Afghanistan, Pakistan, South and Central Europe, Russia and newly independent countries of the former Soviet Union [23, 24]. Two morphological forms of *Anopheles superpictus* in the adult stage in addition to the presence or absence of dark spot on the end of palps female, in terms of the length of the bright spot on the end of palps, wing length and the distance the branching point of 3th or 4th veins to the end of the wing are significantly different. These two forms sympatrically (simultaneously) exist in many parts of the country in terms of geographical distribution. According

to the study of Shemshad et al., out of 168 *Anopheles superpictus* belonging to 10 provinces, 47 (28%) had a spot or terminal dark ring (form B) and 120 (72%) lacked spot (form A) [24]. According to the molecular studies, three genotypes X, Y and Z from *Anopheles superpictus* have been found in Iran. Genotypes Z and Y are sympatrically distributed in Sistan-Baluchistan Province and the genotype X is different in terms of geographical distribution (Allopatric) and is spread in the North, South, West and the Center of the country [23].

There is no documented scientific reports about the difference between the two morphological forms of this species on the behaviors of rest and blood feeding, resistance to insecticides, abundance, power of malaria transmission, geographical distribution, the type of breeding places and adult resting sites, anthropophilic index and its other bionomic characteristics. In the present study, *Anopheles superpictus* had a great relative abundance. This is a normal condition due to the studied area not being located under the pressure of larviciding and spraying and because of plentiful and proper larval habitats and resting places.

In the study of Farsan County and Kurdistan Province, *Anopheles superpictus* respectively consisted 79% and 49.8% of the anopheline mosquitoes [20, 25]. Anopheline mosquitoes fauna in different regions of the country is not similar in terms of species composition and relative abundance due to the climatic difference. In the study of fauna of anopheline species in Jiroft County, eight *Anopheles* were caught, among which *A. stephensi*, *A. pulcherrimus* and *A. fluviatilis* respectively with 63.4%, 17.9% and 10.8% had the highest abundance.

*A. culicifacies*, *A. dthali*, *A. superpictus*, *A. turkhudi* and *A. sergenti* were respectively in later stages [26].

In the survey of *Anopheles* fauna in Guilan Province, a total of eight species were collected, among which the greatest abundance was related to *A. maculipennis* complex (77.45%) and *A. pseudopictus* (15.61%). *A. hircanus*, *A. claviger*, *A. superpictus* and *A. plumbeus* were in the later stages [13]. In the study of 63 larval habitats of Iran Shahr county, 8 species of *Anopheles* including *A. culicifacies*, *A. stephensi*, *A. dthali*, *A. hircanus*, *A. superpictus*, *A. turkhudi*, *A. multicolor* and *A. pulcherrimus* were identified, among which the greatest abundance was related to *A. culicifacies* [27].

*Anopheles dthali*, one of the vectors of malaria in the country, exists in the southern parts of Zagros Mountain ranges [28]. This species is found in provinces of Hormozgan, Baluchistan, Kerman, Fars, Yazd, Khuzestan, Kermanshah, Khorasan and Persian Gulf coastal areas as high as 1410 meters above sea level [29]. *Anopheles dthali* is dispersed in northern Ethiopia and Somalia, Socotra, North Africa to North West Pakistan, South-western Saudi Arabia, around the Red Sea and Aden Gulf. This species is a probable vector in Saudi Arabia and secondary vector of malaria in North Somalia [30]. This *Anopheles* is a secondary vector in some southern parts of the country; especially in the mountainous areas of Hormozgan province [29].

*A. dthali* is found in outdoor places, stables, animal sites and tents. During the day, it rests in water storage tanks, warehouses, cavities and mountain caves, well walls and cavities along rivers. Their adults are very sensitive to light and are often stimulated by flashlight and fly. Anthropophilic index of this species has been reported 1% (Izeh) to 25% (Bandar Abbas) depending on the type of region [28, 30]. Given the exophilic tendencies, *A. dthali* has special importance in terms of maintaining malaria in infected areas. Also, given its more attention to animal bait compared with human bait, the need for research regarding determination of anthropophilic and zoophilic indices is discussed in the under study region.

In the studied areas, anopheline fauna is active from the first half of May to the first half of November and has a peak of activity in the first half of August until the second half of August. The knowledge about the time of maximum activity of malaria vector species can help to combat against anopheline larvae when epidemics occur. In Farsan County, seasonal activity of the dominant species of the region (*A. superpictus*) began from mid-May and continued until mid-October. Second half of July was the peak of activity of this species [20].

The studies conducted in Hormozgan Province showed that the predominant species of region (*A. stephensi*) is active all year round. The activity of this species began in mid-April and had two peaks; one in mid-May to early June and the other one in mid-September to early October (mountainous areas) and mid-October to early November (lowland and coastal areas) [31]. In this province, *A. dthali* is active all year round with two period of abundance; one in mid-September to mid-November and the other one in mid-April to mid-May [30]. The cause of

this difference in the curve of *Anopheles* seasonal activity in different areas is their quite different ecological, weather, climate and topographical conditions.

In this study, seven types of larval habitats were observed, five types of which were natural larval habitats and the two other types were artificial larval habitats. Natural larval habitats including rivers side, streams edge, grasslands, springs and wetlands were respectively 27.5%, 22.9%, 11.8%, 11.8%, 2% and artificial larval habitats including rice crop plots and irrigation canals of farms that respectively consist of 22% and 2% of regional larval habitats.

Locations of larval nests in the *A. superpictus* were rivers side, streams edge, rice crop plots, springs, and wetlands. It laid eggs in larval habitats with clear water, sunny, with or without plants, muddy or sandy bed, permanent or temporary and current or stagnant. Larval habitats of *A. dthali* were streams edge, rice crop plots, rivers side, grasslands, springs and irrigation canals of farms. This species laid eggs in the larval nests with clear water, sunny, with plants or without plants, muddy or sandy bed, permanent or temporary and current or stagnant. In the study of larval nests of mosquitoes in the County of Rasht, out of 35 larval nests (including 8 artificial larval nests and 27 natural larval nests), a total of 44% of larvae were collected from artificial larval nests and 56% were collected from natural larval nests. Larval nests of the dominant species of the region (*A. maculipennis* complex) included rice crop plots or wetlands with herbaceous vegetation, in sun or shade with muddy beds and clear water [19].

In the study of Gilan Province, most anopheline larvae were collected from natural larval habitats (86.9%), such as pits of rivers bed (46.4%) and rain water pits (33.1%) with temporary water (98.3%), stagnant water (99.5%) and clear water (95.3%), plant eligible (69.9%), muddy bottom (42%) or rocky bottom (39.7%), sunny (69.9%), or shade (22.7%) [32]. Comparison of the present study results with these mentioned researches suggests the differences or similarities in the characteristics of larval habitats. Given the biology, species composition and ecological conditions in different regions, these similarities or differences can be largely explained. Macan reports that the majority of larval nests of *A. superpictus* in West Iran to be rocky bed rivers sides. In addition, he reported breeding places of this species to be clear in sunny or semi-shade areas [33].

In Minab county, larval nests of *A. superpictus* were in rivers sides (80%), with stagnant water (62.5%), temporary water (100%), without plants (50%), semi-shade (56.3%) with a muddy bed (56.2%) [34]. The results of these two studies are almost consistent with the present study. Patton reported larval habitats of *A. dthali* to be wells and springs [35]. Manouchehri et al. collected *A. dthali* larvae from mineral water and highly salty water (2.7 parts in 1000). In Hormozgan Province, larval nests of the mentioned species have been reported to be rocky rivers side, springs, spring marginal holes with or without plants, river bed pits and palm irrigation canals [28]. In this study, as previously mentioned, larval habitats of *A.*

*dthali* were observed in rivers side, river bed pits, grasslands, streams edge and rice crop plots.

*A. superpictus* complex is the dominant *Anopheles* species in Luristan Province. Therefore, it is recommended that in future, ecological, biological, cytological and molecular-cellular studies should be conducted in different populations of this complex. In addition, it is recommended that some researches should be conducted to determine behavior, habits as well as infection with arboviruses and dirofilaria in different populations of this species.

One of the limitations of this study is passing relatively great time from conducting this study. However, since no study is conducted in this areas at this time, and several new records were found for the region and also the study in the deprived areas with impassable regions were conducted over a year of long which is very important in terms of malaria transmission and can be a reference for future researches and can be used by those involved in the combat against malaria, so it is extremely important.

In Luristan Province, counties of Khorram-Abad and Aligudarz have a greater potential for malaria transmission and the villagers and tribes in these areas are

at higher risk. Given that these areas have suitable resting places for adults and abundant larval nests for the regional main vector (*A. superpictus*), considering the habits of exophilic and exophagic of this species, education of how to use insecticide-impregnated nets, proper larviciding operations, screening and full treatment of patients, especially in remote mountainous areas are recommended.

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### Authors' Contributions

All authors had role in design, work, statistical analysis and manuscript writing.

### Conflict of Interest

The authors declare no conflict of interest.

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