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Species Composition and Seasonal Dynamics of Mosquito Larvae (Diptera: Culicidae) in Iğdır Plain, Turkey [1]

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Summary

A total of 24.752 larvae consisting of fourteen mosquito species (*Aedes vexans, Anopheles hyrcanus, An. maculipennis s.s., Culex deserticola, Cx. laticinctus, Cx. martinii, Cx. mimeticus, Cx. modestus, Cx. pipiens, Cx. theileri, Cx. territans, Cx. tritaeniorhynchus, Ochlerotatus caspius* and *Oc. dorsalis*) were collected from permanent and temporary breeding areas in Iğdır plain (Ararat Valley) from July through October, 2005 and April through October, 2006 periods. The most dominant species was *Oc. dorsalis* (33.69% of total catch) followed by *An. maculipennis s.s.* (23.02%), *Ae. vexans* (15.19%), *Cx. theileri* (11.3%), *Oc. caspius* (10.33%) and *Cx. pipiens* (2.25%). Except *Oc. caspius,* all of the species could be sampled in permanent breeding areas. On the other hand, only five species (*Oc. dorsalis, Ae. vexans, Oc. caspius, Cx. theileri* and *An. maculipennis* s.s.) were found in temporary breeding areas. The ratio of *An. sacharovi* in Maculipennis complex was 6.5% in Iğdır plain.

Keywords: *Mosquito larvae, Anopheles maculipennis, Anopheles sacharovi, Malaria, Iğdır Plain*

Iğdır Ovası'ndaki (Türkiye) Sivrisinek Larvalarının (Diptera: Culicidae) Tür Kompozisyonu ve Mevsimsel Dinamizmleri [1]

Özet

Temmuz-Ekim 2005 ve Nisan-Ekim 2006 süresince, Iğdır Ovası'nda (Aras Vadisi) kalıcı ve geçici üreme alanlarından 14 sivrisinek türüne (*Aedes vexans, Anopheles hyrcanus, An. maculipennis s.s., Culex deserticola, Cx. laticinctus, Cx. martinii, Cx. mimeticus, Cx. modestus, Cx. pipiens, Cx. theileri, Cx. territans, Cx. tritaeniorhynchus, Ochlerotatus caspius* ve *Oc. dorsalis*) ait 24.752 larva toplandı. En dominant türün *Oc. dorsalis* (toplam yakalananların %33.69'u) olduğu, bu türü, *An. maculipennis* s.s. (%23.02), *Ae. vexans* (%15.19), *Cx. theileri* (%11.3), *Oc. caspius* (%10.33) ve *Cx. pipiens*'in (%2.25) takip ettiği belirlendi. *Oc. caspius* hariç, bütün türler, kalıcı üreme alanlarında örneklenebildi. Diğer taraftan, geçici üreme alanlarında yalnız 5 tür (*Oc. dorsalis, Ae. vexans, Oc. caspius, Cx. theileri* ve *An. maculipennis* s.s.) örneklenebildi. Iğdır Ovası'nda, *An. sacharovi*'nin, Maculipennis complex içerisindeki oranının %6.5 olduğu belirlendi.

Anahtar sözcükler: *Sivrisinek larvası, Anopheles maculipennis, Anopheles sacharovi, Sıtma, Iğdır Ovası*

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INTRODUCTION

Turkey is the last country in the temperate climate zone on the edge of the European continent in which certain vector-borne diseases are prevalent at endemic and occasionally epidemic proportions [1]. With 50 endemic species (10 *Anopheles*, 3 *Aedes*, 15 *Ochlerotatus*, 13 *Culex*, 6 *Culiseta*, 1 *Coquillettidia*, 1 *Orthopodomyia* and 1 *Uranotaenia*), including vectors of malaria and filariasis, mosquitoes are of public health, nuisance and economic importance in much of Turkey. They affect many economically important coastal and inland regions of the country [2].

Malaria was eradicated in all countries of the European Region in the early 1960s except some parts of Azerbaijan, Tajikistan and Turkey, where malaria transmission continued to persist and residual foci of malaria existed [3]. During 1990s, malaria cases increased in Turkey and the Trans-Caucasian countries. The malaria situation in border areas of the above-mentioned countries remains serious [4].

Iğdır plain in Ararat Valley borders with Armenia, Nakhichevan (Autonomous Republic of Azerbaijan) and Iran. Because of appropriate climatic factors, bad drainage system, high underground water level and high salinity, the mosquitoes are represented with high population in Ararat Valley where malaria is endemic.

Detailed information on biology and ecology of the mosquito fauna in the region is necessary for the development of ecologically sensitive and effective mosquito control strategies. The present study established the breeding habitats, population size, seasonal occurrence and abundance of all mosquito species that were necessary for integrated mosquito control activities in the study area.

MATERIAL and METHODS

Study Area

Iğdır plain is the most important irrigated agricultural area in North-eastern Anatolia Region of Turkey. Irrigation water is supplied from River Aras. The plain has salty soil because of alluvium carried by Aras River and materials from volcanic Ararat Mountain. It is 850 meters high from the sea level. Previously cotton was main agricultural product in this area but in present fruit, sugar beet, wheat, barley, leguminous seeds and various vegetables are cultivated. There are numerous large and small drainage canals built by DSI (Public Waterworks Administration) in the plain. These permanent drainage canals and temporary standing water are quite suitable for mosquito breeding.

Iğdır plain is located in the North-eastern Anatolian Region, coldest region of Turkey. The plain is hotter and it has lower rainfall when it is compared to the region, in which it is located. According to the average climatic data of 15 years between 1990 and 2004 [5], the annual average temperature was 9.9°C; the highest temperatures were in July (26°C) and August (25.6°C) and the lowest temperatures was in January (-2.7°C). The annual total rainfall was 250 mm. The highest rainfall was in April (41 mm) and May (50.6 mm); the lowest was in August (7.1 mm). The annual average relative humidity was 51.8 mm.

Selection of Sampling Stations

This study was performed in the area of 400 km^2 in Iğdır plain. Existence of mosquito breeding areas with different characteristics in the study area were taken into consideration for selection of sampling stations [2,6,7]. Thirty-one stations (19 permanent, 12 temporary) were selected for larvae /pupae sampling. Twelve of permanent breeding areas were drainage canals and 7 of them were pools *(Figure 1)*. Temporary habitats in the area consist of depression areas, ponds, pools, small drainage canals, ditches, pasture etc., in which drainage water was accumulated.

Sampling of Mosquito Larvae

Immature stages of mosquitoes were sampled from July through October, 2005 and from April through October, 2006 periods. Samplings were performed by dipping with a standard 400 ml dipper. At least five dipper samples were taken from each larval breeding habitat. The contents of each dipper were transferred into different container and transported to the laboratory. The immature were counted and categorized into 1^{st}-2^{nd} instar, 3^{rd}-4^{th} instar and pupae. The 3^{rd} and 4^{th} instar larvae were identified immediately with the use of keys [8-11], while the 1st and 2^{nd} instar were reared to 3^{rd} and 4^{th}. Pupae were identified after being mature. Some of the larvae were reared to adults in order

Fig 1. The study area and location of mosquito breeding sites [permanent breeding sites (□: drainage canals, O: pools), and temporary breeding sites (Δ)]

Şekil 1. Çalışma alanı ve sivrisinek üreme alanlarının yeri [kalıcı üreme alanları (□: drenaj kanalları, O: su birikintileri) ve geçici üreme alanları (Δ)]

to confirm identification. Sampling procedure was performed twice a month.

Determining the Density Classes of Mosquito Species

The density (relative abundance) of mosquito species in the area was calculated according to the following formula:

D=l/L.100%

Where

D-density, l-number of specimens of each mosquito species, L-number of all specimens, the following density classes were used [12,13]:

Dominant species (D>5%)
Subdominant species (1<D<5%)
Satellite species (D<1%).

Determining *An. sacharovi* in the Study Area

In order to determine whether *An. sacharovi* exists in Iğdır plain, more than 1000 Maculipennis complex specimens, which were fed, were collected by mouth aspirators during July-August 2006 period from animal barns in Kuzugüden, Zülfikar and Melekli villages in the area. These specimens laid eggs. The patterns on the eggs were examined by dissection microscope, and the species were determined in Maculipennis complex [11].

RESULTS

Mosquito Species and Their Relative Abundance

Fourteen mosquito species [*Aedes vexans* (Meigen), *Anopheles hyrcanus* (Pallas), *Anopheles maculipennis* s.s. Meigen, *Culex deserticola* Kirkpatrick, *Culex laticinctus* Edwards, *Culex martinii* Medschid, *Culex mimeticus* Noe, *Culex modestus* Ficalbi, *Culex pipiens* L., *Culex theileri* Theobald, *Culex territans* Walker, *Culex tritaeniorhynchus* Giles, *Ochlerotatus caspius* (Pallas) and *Ochlerotatus dorsalis* (Meigen)] were found in the area.

The most dominant species was *Oc. dorsalis* (33.69% of total catch) followed by *An. maculipennis* s.s. (23.02%), *Ae. vexans* (15.19%), *Cx. theileri* (11.3) and *Oc. caspius* (10.33%). According to density criteria, these mentioned five species were within the dominant class, three species (*Cx. pipiens Cx. modestus* and *Cx. martinii*) were included in subdominant class, and six species (*Cx. laticinctus, Cx. mimeticus, Cx. deserticola, Cx. territans, Cx. tritaeniorhynchus* and *An. hyrcanus*) were in satellite class *(Table 1)*.

Habitat Preferences of Mosquito Species

From permanent breeding sites, 40.79% of 24.752 larvae/pupae (n=10.097) were collected and 59.21% of them (n=14.655) were collected from temporary breeding sites. More than 90% of specimens, collected from permanent sites, belong to four species. These species were *An. maculipennis* s.s. (55.92%), *Cx. theileri* (25.80%), *Cx. pipiens* (5.52%) and *Cx. modestus* (3.50%). On the other hand, only five species from temporary sites could

Table 1. *Mosquito species and their relative abundance (%) in Iğdır plain*
Tablo 1. *Iğdır Ovası'ndaki sivrisinek türleri ve bolluk yüzdeleri*

Mosquito Species	The number of specimens	Relative abundance (%)	The status of the species according to density criteria
Oc. dorsalis	8.338	33.69	Dominant species
An. maculipennis s.s.	5.698	23.02	"
Ae. vexans	3.759	15.19	"
Cx. theileri	2.797	11.30	"
Oc. caspius	2.557	10.33	"
Cx. pipiens	557	2.25	Subdominant species
Cx. modestus	354	1.43	"
Cx. martinii	284	1.14	"
Cx. laticinctus	158	0.64	Satellite species
Cx. mimeticus	88	0.36	"
Cx. deserticola	83	0.33	"
Cx. territans	36	0.14	"
Cx. tritaeniorhynchus	33	0.13	"
An. hyrcanus	10	0.04	"
Total	24.752	100	

Table 2. *The relative abundance (%) of mosquito species in permanent and temporary habitats in Iğdır plain*
Tablo 2. *Iğdır Ovası'nda, kalıcı ve geçici habitatlardaki sivrisinek türlerinin bolluk yüzdeleri*

Mosquito species	Characteristics of habitats									
	Permanent breeding areas						Temporary breeding areas		The contribution of permanent and temporary breeding sites to the populations of the species (%)	
	Drainage canals		Pools		The total number of mosquito in permanent habitats (Drainage canals + pools)		Every kind of water bodies (arable field, pasture, depression areas, ditches, pond, small drainage canal etc.) formed by drainage water generally		Perm.	Temp.
Ae vexans	112*	(1.62)**	44	(1.37)	156	(1.55)	3.603	(24.59)	4.15	95.85
An. hyrcanus	0	(0)	10	(0.31)	10	(0.01)			100	0
An. macul. s.s	3.787	(54.94)	1.859	(58.02)	5.646	(55.92)	52	(0.35)	99.09	0.91
Cx. deserticola	83	(1.21)	0	(0)	83	(0.82)			100	0
Cx. laticinctus	0	(0)	158	(4.93)	158	(1.56)			100	0
Cx. martinii	224	(3.25)	60	(1.87)	284	(2.81)			100	0
Cx. mimeticus	88	(1.28)	0	(0)	88	(0.87)			100	0
Cx. modestus	292	(4.24)	62	(1.94)	354	(3.50)			100	0
Cx. pipiens	478	(6.93)	79	(2.47)	557	(5.52)			100	0
Cx. theileri	1.787	(25.92)	818	(25.53)	2.605	(25.80)	192	(1.31)	93.13	6.87
Cx. territans	0	(0)	36	(1.12)	36	(0.36)			100	0
Cx. tritaenior.	24	(0.35)	9	(0.28)	33	(0.33)			100	0
Oc. caspius	0	(0)	0	(0)	0	(0)	2.557	(17.44)	0	100
Oc. dorsalis	18	(0.26)	69	(2.15)	87	(0.86)	8.251	(56.30)	1.04	98.96
Total	6.893	(68.27)	3.204	(31.73)	10.097	(100)	14.655	(100)		

* Number of specimens ** Relative abundance of the species

be sampled. Larvae belonging to *Oc. dorsalis* (56.30%), *Ae. vexans* (24.59%) and *Oc. caspius* (17.44%) consist of 98.33% of the larvae collected from temporary sites. The ratios of *An. maculipennis* s.s. and *Cx. theileri* were very low (0.35% and 1.31%, respectively) in temporary sites, while they were the most dominant species in permanent sites *(Table 2)*.

From permanent sites 6.893 of larvae/pupae (68.27%) were collected from drainage canals and 3.204 (31.73%) were collected from pools. Drainage canals and pools had different characteristics from the point of the species and their relative abundance. That is why, two species (*Cx. deserticola* and *Cx. mimeticus*) were only sampled from drainage canals and three species (*An. hyrcanus Cx. laticinctus and Cx. territans*) were only sampled from pools *(Table 2)*. *An. maculipennis* s.s. (n=3.787 in drainage canals and n=1.859 in pools) and *Cx. theileri* (n=1.787 in drainage canals and n=818 in pools), which were the most dominant species in permanent sites, were sampled more in drainage canals than in pools. However, for both species there was no significant difference between drainage canals and pools (P>0.05). Larvae number of *Cx. pipiens, Cx. martinii,* and *Cx. modestus* (n=478, n=224 and n=292 respectively) sampled from drainage canals were more than the larvae number of these species sampled from pools (n=79, n=60 and n=62 respectively). For these three species there was significant difference between drainage canals and pools (P<0.05).

It was determined that the contribution of permanent and temporary sites to the species population density was quite different. All of the specimens of 9 species (*An. hyrcanus, Cx. deserticola, Cx. laticinctus, Cx.martinii, Cx. mimeticus, Cx. modestus, Cx. pipiens, Cx. territans* and *Cx. tritaeniorhynchus*), 99.09% of *An. maculipennis* s.s. and 93.13% of *Cx. theileri* were collected from permanent breeding sites. On the other hand, all of the specimens of *Oc. caspius*, 95.85% of *Ae. vexans* and 98.96% of *Oc. dorsalis* were collected from temporary breeding sites *(Table 2)*.

While the number of species was high (13 species) and the number of specimens was low in permanent sites, the number of species was low (5 species) and the number of specimens was high in temporary sites, except *An. maculipennis* s.s. and *Cx. theileri (Table 2)*.

It was found that the number of species in both drainage canals and pools, which were permanent sites, were high. In some stations (11, 17) six species and in some stations (5, 18) seven species were sampled together. *An. maculipennis* s.s. was sampled in all of 19 permanent sites, and *Cx. theileri* was sampled in 16 of 19 permanent sites. In some of temporary sites (2, 4 and 12) three species were sampled together.

Monthly Changes in Population Fluctuations of Mosquito Species

An. maculipennis s.s., which could be sampled continuously, reached its highest population density during the months of July and August, 2005 (n=1.765 and n=615 respectively) and June-July, 2006 (n=908 and n=1.120 respectively) periods. After these periods regular decrease was determined in the population density of the species. *An. hyrcanus* was sampled only in October, 2005 period with low number (n=10).

The larvae of *Cx. theileri* and *Cx. pipiens* were collected during all of the periods except April 2006. *Culex theileri,* which had high population density, having regular population fluctuations, reached its peak in both July-August, 2005 (n=614 and n=541 respectively) and July-August, 2006 periods (n=321 and n=526 respectively). Although the population density of *Cx. pipiens* increased in September 2005 period (n=118), during the following periods of the study there was no notable peak in population density of the species. The *Culex* species in the area that had low population density (*Cx. deserticola, Cx. laticinctus, Cx. martinii, Cx. mimeticus, Cx. modestus, Cx. territans* and *Cx. tritaeniorhynchus*) had similarities in population fluctuations. None of these could be sampled continuously and the larvae of most of these species could not be collected after second part of summer *(Figure 2)*.

Aedes vexans, Oc. caspius and *Oc. dorsalis* could be sampled from May until October (five month period). Despite the irregular fluctuations of the population of *Oc. caspius,* it reached its peak in August, 2006 period (n=1.375). While *Ae. vexans* reached its peak in August (n=1.318 in August, 2005 and n=991 in August, 2006), *Oc. dorsalis*, which was the most dominant species in the area, reached its peak during August, 2005 (n=1.213) and July- August, 2006 periods (n=2.602 and n=1.982 respectively) *(Figure 2)*.

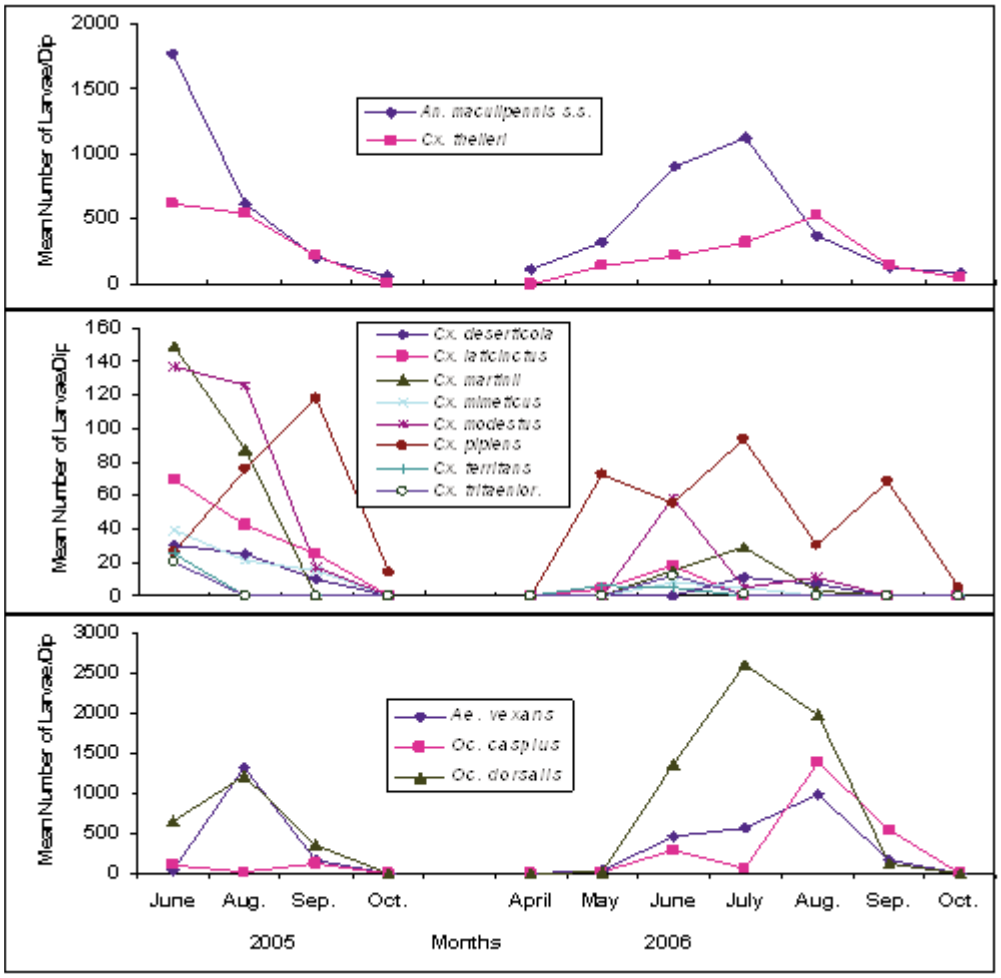

Fig 2. Monthly population fluctuations of mosquito larvae in Iğdır plain during the study period

Şekil 2. Çalışma peryodu süresince, Iğdır Ovası'nda sivrisinek larvalarının aylık populasyon dalgalanmaları

The Ratio of *An. sacharovi* in Maculipennis Complex

Total 523 specimens belonging to Maculipennis complex laid eggs. The ratio of *An. sacharovi* in Maculipennis complex was 6.5% (34/523). The results from different villages were given in *Table 3*.

Table 3. *The ratio of An. maculipennis and An. sacharovi in Maculipennis complex, collected from three villages in Iğdır plain*
Tablo 3. *Iğdır Ovası'ndaki üç köyden toplanan Maculipennis complex içerisinde An. maculipennis ve An. sacharovi'nin oranları*

Village	The number of Maculipennis complex specimens	The number of *An. maculipennis*	The number of *An. sacharovi*
Kuzugüden	107	107 (100%)	0 (0.00%)
Zülfikar	196	183 (93.37%)	13 (6.63%)
Melekli	220	199 (90.46%)	21 (9.54%)
Total	523	489 (93.5)	34 (6.5%)

DISCUSSION

The existence of *Anopheles maculipennis* s.s. in all of 19 permanent breeding sites and high relative abundance (55.92%) in these sites shows that the species has high adaptation to the Iğdır plain. *An. maculipennis* was the most dominant species in Armenia which borders with our study area [14]. After malaria outbreaks in Armenia in recent years, the importance of reappearance of *An. sacharovi* in the country was emphasized [15]. In this study, *An. sacharovi*, the most important malaria vector in Turkey, was found in the area and the ratio of this species was 6.5% in Maculipennis complex. In Transcaucasia Ecological Region, which also includes Iğdır plain, *An. maculipennis* and *An. sacharovi* with some other *Anopheles* species are important malaria vectors [16,17]. We found that *Anopheles hyrcanus* had the lowest population density in Iğdır plain. In a study performed with

light traps, it was determined that the density of *An. hyrcanus* was low (0.7% of total catch) in Iğdır plain [18]. According to Ramsdale [19], some forms of *An. hyrcanus* s.l. are effective malaria vectors in east and south-east Asia.

Culex theileri was sampled during study period, except April 2006 *(Figure 2)*. It was determined that the species had high distribution rate (84.21%, 16 of 19 sites) in permanent breeding sites. A study was performed in Gölbaşı district of Ankara province and in the study it was determined that *Cx. theileri* was associated with seven other species in different habitat types, such as marsh, lake shore, creek side, canal, animal footprints, pasture and waste water pool [20]. According to Harbach [9], *Cx. theileri* is an adaptive species which utilizes a wide range of breeding sites, but it is also a focal species, reaching high densities in some areas while appearing to rare or absent in others. In Şanlıurfa province in southeastern part of Turkey, which has similar ecological characteristics to our study area, it was found that *Cx. theileri* had high population density and it could adapt to different breeding sites [20]. In the same study it was found that *Cx. martinii* and *Cx. territans* had lower population density than *Cx. pipiens* and *Cx. theileri*.

Culex pipiens, which was in subdominant species category in Iğdır plain (2.25% of total catch), could only be sampled in 3 permanent sites and it had low distribution rate (15.79%). Harbach [9] reported that the species had high population densities generally in habitats in urban regions, rather than rural regions. While the rates of *Cx. theileri* and *Cx. pipiens* were 9.44% and 12.05%, respectively, collected by light traps in study area [18], these rates were 9.25% and 1.49% according to biting activities results [21]. Reinert [22] reported that certain differences in relative abundance of mosquito species can be found in studies which are carried out by different sampling methods. *Culex pipiens* adults feed almost exclusively on birds, and this species is one of the most important vectors of West Nile virus (WNV) [10,23]. It is very important for Ararat Valley. This importance results from some factors: in the area, which is located on bird migration route, 190 bird species have been observed, and the Valley also consist of different characteristic wetlands, and the mosquitoes are represented in high populations [24,25]. Although WNV could not be detected in mosquito species, the virus was found in human serum samples in the studies performed in different regions of Turkey [26,27].

We detected that *Ochlerotatus dorsalis* (33.69% of total catch), the most dominant species in the study area, had very high larval density in some temporary breeding sites (500-600 larvae/dip). Gündüz [18] reported that *Oc. dorsalis* was the most dominant species (72.13% of total catch) caught by light traps in Iğdır plain. We collected 59.21% of 24.752 larvae (n=14.655) from temporary breeding sites. The larvae belonging to *Oc. dorsalis* (56.30%), *Ae vexans* (24.59%) and *Oc. caspius* (17.44%) collected from temporary sites consist of 98.33% of specimens in the temporary sites *(Table 2)*. *Ochlerotatus dorsalis* were collected from different geographic areas such as lagoons, salty marshes, salty pools, spring pools, snow pools, shallow irrigation seeps, pools on flooded plains, irrigation and artesian water [28]. According to Snow [10], the breeding areas of *Oc. dorsalis* are sun lighted, or partially shaded, temporary collections of fresh or brackish water in ditches and pools; the egg-laying sites of *Oc. caspius* are usually coastal marshes although there are many records from inland, non saline waters.

Temporary breeding sites in Iğdır plain were originated from irrigation and drainage water. The larvae of *Aedes vexans*, *Oc. caspius* and *Oc. dorsalis* consist of 98.33% of larvae, which we collected from temporary sites *(Table 2)*. In a study performed in Çukurova on Mediterranean coast, *Oc. caspius* was highly sampled from temporary breeding sites, which was formed by drainage water [29]. The most characteristic feature of larval site for *Ae. vexans* is fresh water, collections of seepage water over grass or sodden areas produce larvae in irrigated areas [28]. These three species (*Ae vexans, Oc. caspius* and *Oc. dorsalis*) are the most important vectors for Tahyna virus, which is present in most countries of central and southern Europe [30]. Although there are no data related to Tahyna virus in the study area, the presence of this virus in Aegean region of Turkey was determined in the population [26]. According to Bedir [21], total relative abundance of *Oc. dorsalis* (47.46%), *Oc. caspius* (4.88%) and *Ae. vexans* (6.57) was 58.91% in the study of biting activity on man in the area.

During the study period, samplings could be done only once from some of the temporary breeding sites, on the other hand, samplings could be done maximum three times from some other temporary sites in a year since the collection of

water in these breeding areas depend on irrigation of surrounding field. However, contribution of these breeding sites to the total mosquito population density in Iğdır plain was 59.21%. It can be caused by the reality that the larvae/dip rate was about a few hundred in temporary breeding sites.

For the mosquito larvae control activities, temporary breeding sites for three species (*Oc. dorsalis, Oc. caspius* and *Ae. vexans*) and permanent breeding sites for *Anopheles* and *Culex* species are very important in Iğdır plain. Besides, high adaptation capability of *An. maculipennis* s.s., which is malaria vector in the area, to the permanent breeding areas should be taken into consideration.

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