

A recent survey of the mosquito (Diptera: Culicidae) fauna and seasonal human biting activity in the city of Chisinau, Moldova

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Abstract: This is the first report on species composition and seasonal human biting activity of mosquitoes in the city of Chisinau, Moldova. In total 3,255 adult mosquitoes were collected in eight recreational areas. Sampling occurred by means of two methods: human landing collection and net-catches from vegetation. Altogether, 22 mosquito species in nine genera were found, representing 55% of the species diversity in Moldova. Seventeen anthropophilic species with different seasonal patterns of biting activity were found in human landing collections. The most frequently captured species was *Aedimorphus vexans* (N = 1,514; 46.5%) followed by *Ochlerotatus annulipes* (N = 403; 12.4%), *Dahlia geniculata* (N = 330; 10.1%), *Culex modestus* (N = 312; 9.6%), *Coquillettidia richiardii* (N = 144; 4.4%) and *Oc. riparius* (N = 117; 3.6%). Eight of the species are the known vectors of West Nile virus in Europe; four of them are the natural vectors of *Dirofilaria*. *Journal of the European Mosquito Control Association* 31: 1-7, 2013

Keywords: Culicidae, anthropophilic mosquitoes, seasonal activity, recreational areas, Chisinau

Introduction

Chisinau is situated in the centre of the Republic of Moldova on the river Bic. The surface area of the city is about 120 sq. km, with a total area of green spaces within the city of -3388.6 ha, and a human population of -593,000 inhabitants. There are 23 permanent bodies of water in Chisinau that are mostly situated in public woodland parks (Colesnic, 1996; Tikhonova *et al.*, 2012). These recreational areas are close to residential complexes and produce large numbers of adult mosquitoes that directly impact the human population. Mosquitoes are nuisance insects, particularly when they occur in mass numbers. Not only can the bites of female mosquitoes trigger allergies caused by introduction of the salivary gland secretions into the human blood, but they are recognized worldwide as important vectors of a wide range of viral and parasitic diseases that affect both humans and animals (Becker *et al.*, 2010).

West Nile, Batai and Tahyna viruses, as well as *Plasmodium vivax*, *P. falciparum* and *P. malariae*, which are transmitted by mosquitoes, have been reported in Moldova (Markovich *et al.*, 1949; Sergeyeva, 1953; Miliutina *et al.*, 1957; Chumakov *et al.*, 1974; Scoferta *et al.*, 1984, 1989, 1996). Serological investigations recorded females of *Anopheles maculipennis* Meigen *s.l.* positive for Batai virus and females of *Culex modestus* Ficalbi positive for arbovirus of the genus Orbivirus in Moldova (Chumakov *et al.*, 1981). West Nile virus was first isolated from *Ixodes ricinus* Fabricius and *Derma-centor marginatus* Sulzer in Moldova (Chumakov *et al.*, 1974). Human cases of *Dirofilaria repens*, which is transmitted from animal to humans by mosquitoes, has also been reported in Moldova (Gudumac *et al.*, 2011).

In Poland and the Czech Republic, studies on the presence of *Borrelia burgdorferi sensu lato* in mosquitoes from recreational areas indicated their possible role in the epidemiology of borreliosis. The risk of acquiring *Borrelia* transmitted by mosquitoes is diminished by the two-week survival of spirochetes in the mosquito host (Hard, 1966; Halouzka, 1993; Kosic-Bogacka *et al.*, 2002).

The species composition of anthropophilic mosquitoes in Chisinau city is poorly known. Only a few studies have been dedicated to *An. maculipennis s.l.* and its control due to the malaria outbreaks in Chisinau at the beginning of the 20th century (Sergeyeva, 1953; Miliutina *et al.*, 1957)

To conduct risk analyses and develop emergency management strategies for arthropod-borne diseases, current information on potential vectors in this area is required. The aim of the present study was to determine the species composition and seasonal activity of anthropophilic mosquitoes inhabiting recreational areas of Chisinau that are potential vectors of mosquito-borne pathogens.

Materials and Methods

Study sites

Surveys were carried out in eight recreational areas and small forest plots within the Chisinau city as follows: Botanical Garden (104 ha), Valea Trandafirilor Park (148 ha), Dendrariu Park (83 ha), La Izvor Park (163.5 ha), Riscani Park (87.1 ha), forest plots in Bacioii Noi and Telecentru. Entomological surveys were performed near the Ghidighici water storage basin as well (Fig. 1). The studied areas consist of deciduous forests, open grassland, permanent water basins and canals. These sites have favourable ecological conditions for the interaction between resident bird reservoirs, migratory bird reservoirs from arbovirus endemic areas in Europe and Africa, as well as ornithophilic and anthropophilic mosquito vectors implicated in enzootic circulation of arboviruses (Reusken *et al.*, 2011; Tibuleac, 2011;). Over 75 different bird species, such as *Anas platyrhynchos*, *Larus ridibundus*, *Crex crex*, *Sturnus vulgaris*, *Corvus frugilegus*, *C. corone*, *C. corax*, *C. monedula*, *Pica pica*, *Garrulus glandarius*, *Phylloscopus collybitus*, *Phasianus colchicus*, *Turdus pilaris*, *Passer domesticus*, *P. montanus*, *Regulus regulus*, *Fringilla coelebs*, *Parus major*, *Hirundo rustica*, *Phoenicurus ochruros*, *Columba palumbus*, etc., and 35 species of mammals like *Muscardinus avellanarius*, *Clethrionomys glareolus*, *Microtus arvalis*, *Arvicola terrestris*, *Apodemus uralensis*, *Ap. sylvaticus*, *Ap. flavicollis*,

Ap. agrarius, *Mus musculus*, *Micromys minutus*, *Rattus norvegicus*, *Sorex minutus* etc., have been recently observed in Chisinau (Toderas *et al.*, 2010; Tibuleac, 2011; Tikhonova *et al.*, 2012).

Mosquito collections

Surveys were conducted every ten days between May and September during 2010, 2011 and 2012, using human landing collections and net-catches from vegetation (Silver, 2008). All mosquitoes landing on human bait were caught using 60x15 mm glass tubes. On each occasion, mosquitoes were collected for 5-30 minutes. Mosquitoes were sometimes collected by slowly walking through vegetation and capturing them in small hand-nets as they are disturbed and take flight. Such collections sample unfed, blood-fed and gravid females, and also males. Resting collections provide much additional information and yield samples of the population that more accurately represent the sex ratio (Silver, 2008).

Mosquito identification

Adult mosquitoes were identified using the keys of Cranston *et al.* (1987) and Becker *et al.* (2010). Females of *Culex pipiens* (*Culex pipiens* Linnaeus biotype *pipiens* and *Cx. pipiens* biotype *molestus* Forskal) and *Cx. torrentium* Martini collected from vegetation were identified as *Cx. pipiens s.l./Cx. torrentium*, but females collected on human bait were identified as *Cx. pipiens s.l.* The males of *Aedimorphus vexans* (Meigen), *Aedes geminus* Peus, *Dahlia geniculata* (Oliver), *Cx. pipiens*, *Cx. torrentium*, *Cx. territans* Walker, *Cx. modestus* Ficalbi, *Culiseta annulata* (Schrank), *Coquillettidia richiardii* (Ficalbi), *Ochlerotatus annulipes* (Meigen), *Oc. caspius* (Pallas), *Oc. sticticus* (Meigen) and *Uranotaenia unguiculata* Edwards, were identified to species on diagnostic male genitalic structures. The systematic arrangement takes into account the taxonomic classification used in the online catalog of the Walter Reed Biosystematics Unit and the Mosquito Taxonomic Inventory.

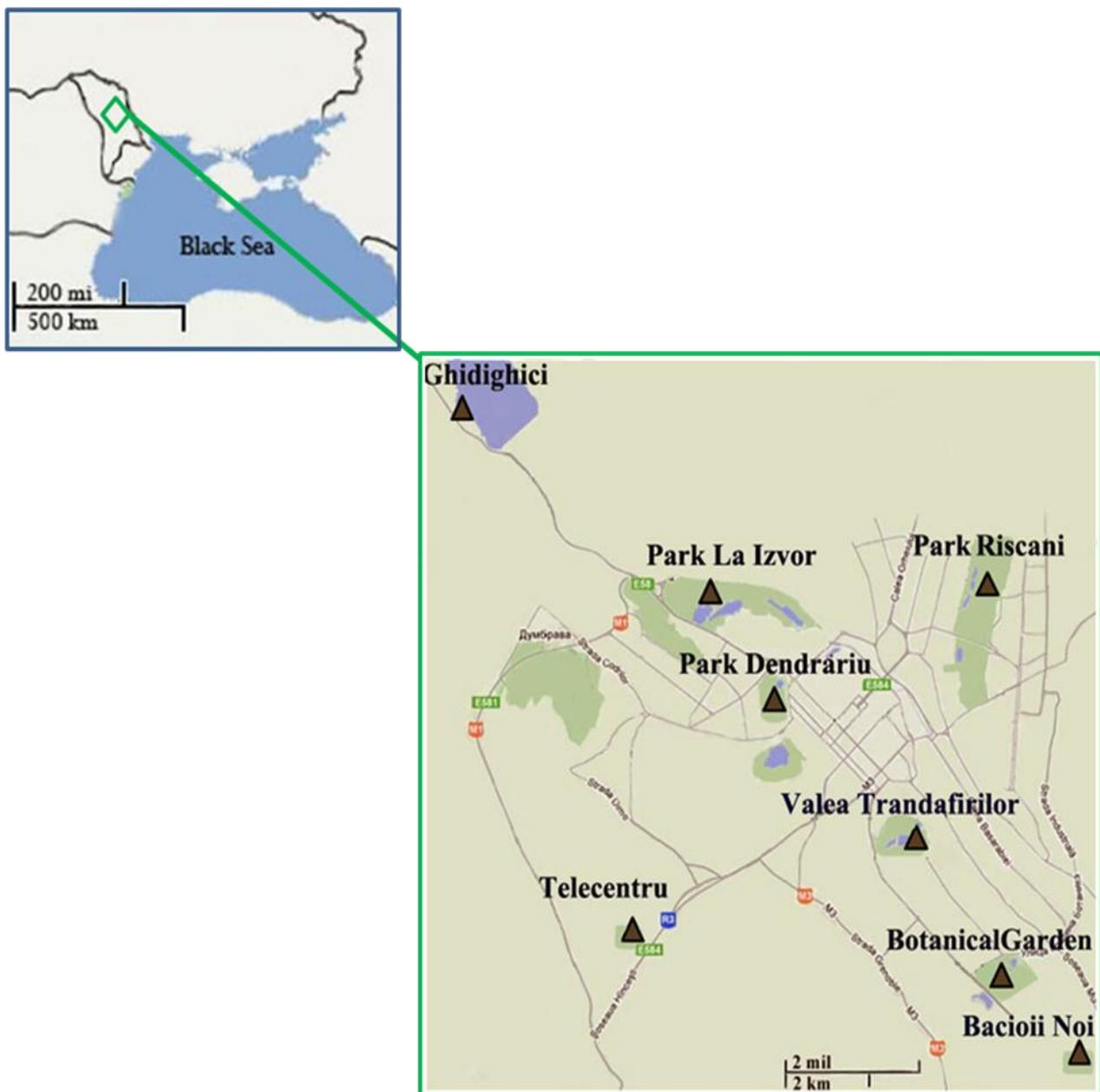


Figure 1. Location of eight recreational areas and small forest plots in Chisinau city where mosquitoes were sampled 2010 – 2012.

Results and Discussion

A total of 3,255 adult mosquitoes belonging to nine genera and 22 species were caught on human bait (Table 1) and in net-catches of resting mosquitoes (Table 2). The most frequently captured species by both methods was *Am. vexans* (N = 1,514; 46.5%), followed by *Oc. annulipes* (N = 403; 12.4%), *Da. geniculata* (N = 330; 10.1%), *Cx. modestus* (N = 312; 9.6%), *Cq. richiardii* (N = 144; 4.4%) and *Oc. riparius* (N = 117; 3.6%). Seventeen anthropophilic species with different patterns of seasonal biting activity have been found in human landing collections (Table 1). Comparative analysis between genera revealed the dominant genus was *Aedimorphus* (48.3%) followed by *Ochlerotatus* (25.6%), *Dahlia* (10.6%), *Culex* (9.9%), *Coquillettidia* (4.9%) and *Anopheles* (0.6%).

Human biting activity of *Am. vexans* during each 10-day period each month was consistent throughout the five months

Table 1. Human biting population of female mosquitoes collected on human bait in the recreational areas of Chisinau city from 2010 to 2012

Mosquito species	May	June	July	Aug	Sept	Total (%)
<i>Anopheles plumbeus</i>	-	15	-	-	3	18 (0.6)
<i>Aedimorphus vexans</i> **	3	534	627	153	96	1413 (48.3)
<i>Dahlia geniculata</i>	2	117	165	27	-	311 (10.6)
<i>Ochlerotatus annulipes</i>	34	114	255	-	-	403 (13.8)
<i>Oc. behningi</i>	-	3	6	-	-	9 (0.3)
<i>Oc. cantans</i> *	12	33	27	-	-	72 (2.5)
<i>Oc. caspius</i> **	-	9	2	-	-	11 (0.4)
<i>Oc. cataphylla</i> *	27	-	6	-	-	33 (1.1)
<i>Oc. communis</i>	7	-	-	-	-	7 (0.2)
<i>Oc. dorsalis</i>	-	4	-	-	2	6 (0.2)
<i>Oc. excrucians</i> *	30	15	27	3	-	75 (2.6)
<i>Oc. pulcritarsis</i>	-	-	3	-	-	3 (0.1)
<i>Oc. riparius</i>	18	45	54	-	-	117 (4.0)
<i>Oc. sticticus</i>	-	6	3	3	-	12 (0.4)
<i>Culex modestus</i> *	2	119	78	30	12	241 (8.2)
<i>Cx. pipiens s.l.</i> **	3	5	12	21	9	50 (1.7)
<i>Coquillettidia richiardii</i> **	-	81	45	18	-	144 (4.9)
Total	138	1100	1310	255	122	2925

* natural vector of West Nile virus; + natural vector of dirofilariæ.

The host-seeking females of *Da. geniculata* were active throughout the first four months of the study, with lowest activity in May and August. The highest human biting activity was during the first 10-day period of July (12–13 mosquitoes per hour) (Figs. 2a&b). This species was not captured in September. It was recorded from five sites with relative numerical abundance from 1.8% to 13%.

Culex modestus was present throughout the six months, with lowest activity in May and September and the highest human biting activity during the third 10-day period of June (17–18 mosquitoes per hour). The species was recorded in six recreational areas with relative numerical abundance from 1.7% to 20.5% (Table 3).

Coquillettidia richiardii was present only in summer from the second 10-day period of June until the third 10-day period of August, with maximum biting activity from the third 10-day period of June until the second 10-day period of July (5–6 mosquitoes per hour). This species was recorded in three recreational areas with relative numerical abundance from 2.9% to 10.2% (Table 3).

of the study (Table 1, Figure 2). The maximal peak of activity was from the third 10-day period of June until the third 10-day period of July (14–22 mosquitoes per hour). This species was recorded from each of eight recreational areas with relative numerical abundance from 9.8% to 76.4% (Table 3).

Studies of 10-day and monthly activity of *Oc. annulipes* showed its presence in May, June and July (Table 1), with the highest biting activity from the third 10-day period of June until the first 10-day period of July (7–12 mosquitoes per hour) (Fig. 2). The species was not present during the hot period of the season. *Ochlerotatus annulipes* was found in six sites with relative numerical abundance from 2.3%–35.3% (Table 3).

Ochlerotatus riparius was active throughout the first three months from the second 10-day period of May until the second 10-day period of July (Fig. 2). The highest human biting activity was recorded during the first 10-day period of July (9–10 mosquitoes per hour). This species was present in four sites with relative numerical abundance from 0.7%–21.6% (Table 3).

Collections of outdoor resting mosquitoes revealed the presence of 13 species, including five species that were not captured on human baits: *Cx. torrentium* (feeds on birds), *Cx. territans* (feeds on amphibians, reptiles and birds), *Ur. unguiculata* (feeds on amphibians), *Ae. geminus* (feeds on mammals and humans) and *Cs. annulata* (feeds on mammals, humans and birds) (Becker *et al.*, 2010). In these collections, *Am. vexans* was the most abundant species (30.6%) followed by *Cx. modestus* (21.5%) and morphologically similar females of *Cx. pipiens* and *Cx. torrentium* (17.6%) (Table 2).

The composition of mosquitoes in the recreational areas are as follow: Valea Trandafirilor Park – 16 species, Botanical Garden – 6 species, Dendrariu Park – 7 species, La Izvor Park – 7 species, Riscani Park – 7 species, Bacioii Noi – 18 species,

Telecentru – 8 species and Ghidighici – 5 species (Table 3). Eight of the species are known vectors of West Nile virus in Europe and four are natural vectors of dirofilariiae (Coluzzi &

Trabucchi, 1968; Cancrini *et al.*, 2006; Heyman *et al.*, 2008; Hubalek *et al.*, 2010).

Table 2. Mosquito species collected from vegetation by entomological nets in the recreational areas of Chisinau city from 2010 to 2011

Mosquito species	May	June	July	Aug	Sept	Total
<i>Aedes geminus</i>	-	1♂	-	-	-	1♂
<i>Am. vexans</i>	-	39♂ 2♀	26♂	5♂13♀	2♂14♀	72♂ 29♀
<i>Da. geniculata</i>	-	7♂ 1♀	11♂	-	-	18♂ 1♀
<i>Oc. behningi</i>	-	1♂	-	-	-	1♂
<i>Oc. caspius</i>	-	-	-	5♂	-	5♂
<i>Oc. excrucians</i>	1♂	-	-	1♀	-	1♂1♀
<i>Oc. sticticus</i>	-	1♂	-	-	-	1♂
<i>Cx. modestus</i>	-	12♀ 21♂	28♂	3♀6♂	1♀	55♂16♀
<i>Cx. pipiens/ Cx.torrentium</i>	-	-	24♀	30♀	4♀	58♀
<i>Cx. pipiens s.l.</i>	-	3♂	17♂	29♂	2♂	51♂
<i>Cx. torrentium</i>	-	3♂	4♂	2♂	4♂	13♂
<i>Cx. territans</i>	-	-	-	2♂ 2♀	-	2♂ 2♀
<i>Culiseta annulata</i>	-	-	-	1♂	-	1♂
<i>Ur. unguiculata</i>	-	-	-	-	2♀	2♀
Total	1	91	110	99	29	330

Table 3. Relative abundance of mosquito species collected in eight recreational areas of Chisinau city from 2010 to 2012.

Species	Location							
	Valea Trandafir. (%)	Botanical Garden (%)	Dendrariu Park (%)	La Izvor Park (%)	Riscani Park (%)	Bacioii Noi (%)	Telecentru (%)	Ghidighici (%)
<i>An. plumbeus</i>	1.2	-	1.8	-	-	-	-	-
<i>Ae. geminus</i>	0.2	-	-	-	-	-	-	-
<i>Am. vexans</i>	54.4	35.9	76.4	9.8	43.2	37.7	22.7	68.2
<i>Da. geniculata</i>	10.7	-	1.8	-	11.4	13.0	10.6	-
<i>Oc. annulipes</i>	4.9	-	-	35.3	2.3	18.1	6.1	19.1
<i>Oc. behningi</i>	0.5	-	-	-	-	0.5	-	-
<i>Oc. cantans</i>	1.9	-	-	9.8	-	2.2	3.0	-
<i>Oc. caspius</i>	0.2	-	1.8	-	-	0.5	-	5.0
<i>Oc. cataphylla</i>	-	-	-	3.9	-	2.2	-	-
<i>Oc. communis</i>	-	-	-	-	-	0.7	-	-
<i>Oc. dorsalis</i>	0.2	-	-	-	-	-	1.5	-
<i>Oc. excrucians</i>	0.5	2.6	-	5.9	-	4.6	1.5	-
<i>Oc. pulcritarsis</i>	-	-	-	-	2.3	0.2	-	-
<i>Oc. riparius</i>	0.7	-	-	21.6	-	4.2	10.6	-
<i>Oc. sticticus</i>	0.7	-	-	-	-	0.5	-	-
<i>Cx. modestus</i>	13.6	8.1	5.5	-	20.5	1.7	-	4.5
<i>Cx. pipiens</i> ♂♀	2.2	15.3	9.1	-	11.4	3.2	39.4	-
<i>Cx. pipiens/Cx. torrentium</i> ♀	-	25.6	-	7.8	-	3.9	19.7	-
<i>Cx. torrentium</i> ♂	0.5	-	3.6	5.9	9.1	0.2	-	3.9
<i>Cx. territans</i>	-	-	-	-	-	1.0	-	-
<i>Cs. annulata</i>	-	-	-	-	-	0.2	-	-
<i>Cq. richiardii</i>	7.5	10.2	-	-	-	2.9	-	-
<i>Ur. unguiculata</i>	-	2.1	-	-	-	-	-	-

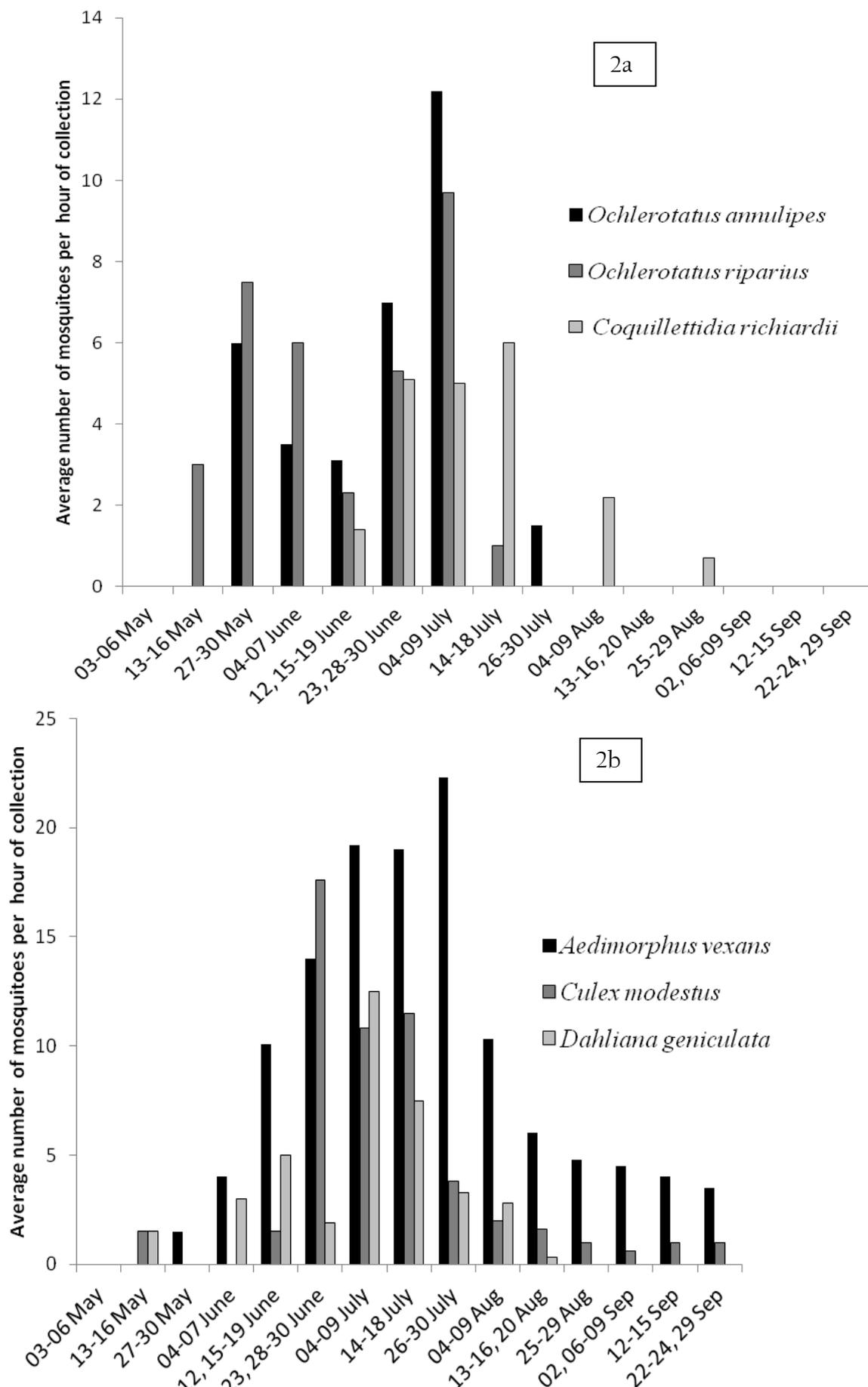


Figure 2a and 2b. Human biting activity of mosquito species captured on human bait in Chisinau city between May and September of each year from 2010 to 2012. The numbers represent the estimated mosquitoes per one hour of collection.

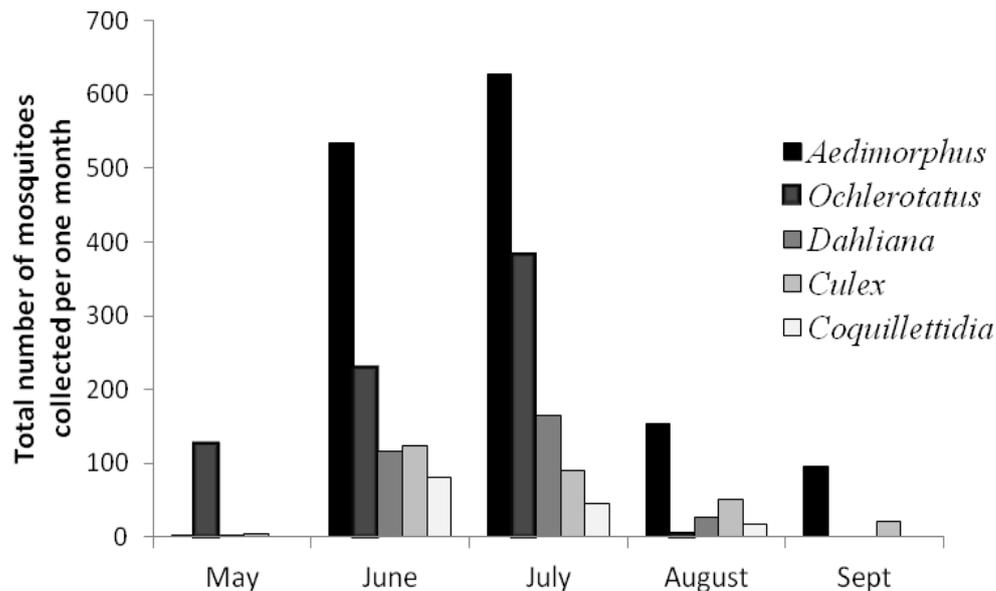


Figure 3. Comparison of genera of mosquitoes in human landing collections in Chisinau city between May and September of each year from 2010 to 2012.

Conclusions

In total, 3,255 adult mosquitoes were collected on human bait and net catches of mosquitoes resting on vegetation in Chisinau. Twenty-two species of nine genera were found, representing 55% of the species known to occur in the Republic of Moldova: *An. plumbeus*, *Ae. geminus*, *Am. vexans*, *Da. geniculata*, *Oc. annulipes*, *Oc. behningi*, *Oc. cantans*, *Oc. caspius*, *Oc. cataphylla*, *Oc. communis*, *Oc. dorsalis*, *Oc. excrucians*, *Oc. pulcritarsis*, *Oc. riparius*, *Oc. sticticus*, *Cx. modestus*, *Cx. pipiens*, *Cx. torrentium*, *Cx. territans*, *Cs. annulata*, *Cq. richiardii* and *Ur. unguiculata* (Sulesco *et al.*, 2012). Seventeen anthropophilic species with different patterns of seasonal biting activity were obtained in human landing collections. The most frequently captured species on human bait were *Am. vexans*, *Oc. annulipes*, *Da. geniculata*, *Cx. modestus*, *Cq. richiardii* and *Oc. riparius*. Their 10-day period of biting activity was studied in detail for five months (May–September). Five additional species, i.e. *Cx. torrentium*, *Cx. territans*, *Ur. unguiculata*, *Ae. geminus* and *Cs. annulata* were captured from vegetation that were not present in human landing collections, probably because of specific host preferences.

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