new species of the Hyrcanus Group based on

PCR techniques in the country. Malaria is

the most important mosquito-borne disease

in Iran and seven species of the genus Ano-

pheles: An. culicifacies Giles s.l., An. dthali

Patton, An. fluviatilis James s.l., An. macu-

lipennis Meigen s.l., An. sacharovi Favre,

An. stephensi Liston, and An. superpictus are

known its proven vectors in Iran, also An.

pulcherrimus Theobald is mentioned a sus-

pected vector of malaria in southeastern Iran

(Edrissian 2006). Eshghy (1977) observed

Plasmodium oocysts in An. multicolor Cam-

bouliu, but sporozoites have not been found

<u>Original Article</u> Mosquito (Diptera: Culicidae) Fauna of Qom Province, Iran

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Abstract

Background: There is very little information about the mosquito fauna of Qom Province, central Iran. By now only three species; *Anopheles claviger*, *An. multicolor*, and *An. superpictus* have been reported in the province. To study mosquito fauna and providing a primary checklist, an investigation was carried out on a collection of mosquitoes in this province.

Methods: To study the mosquito (Diptera: Culicidae) fauna, larval collections were carried out from different habitats on 19 occasions using the standard dipping technique during spring and summer 2008 and 2009.

Results: In total, 371 mosquito larvae were collected and morphologically identified including 14 species representing four genera: Anopheles claviger, An. marteri, An. turkhudi, An. superpictus, Culex arbieeni, Cx. hortensis, Cx. mimeticus, Cx. modestus, Cx. pipiens, Cx. territans, Cx. theileri, Culiseta longiareolata, Cs. subochrea, and Ochlerotatus caspius s.l. All species except for An. claviger and An. superpictus were collected for the first time in the province. All larvae were found in natural habitats. The association occasions and percentages of the mosquito larvae in Qom Province were discussed.

Conclusion: There are some potential or proven vectors of different human and domesticated animal pathogens in Qom Province. The ecology of these species and the unstudied areas of Qom Province need to be investigated extensively.

Keywords: Anopheles, Culex, Culiseta, Ochlerotatus, larvae, taxonomy

Introduction

According to the most recent classification of mosquitoes, the family Culicidae (Diptera) includes two subfamilies, 11 tribes, 111 genera, and 3523 species in the world fauna and the genus *Anopheles* Meigen includes seven subgenera and at least 464 species (Harbach 2007). The mosquito fauna of Iran includes seven genera, 64 species, and 3 subspecies and the records of 12 other species need to be verified (Azari-Hamidian 2007). Recently, Oshaghi et al. (2008) introduced *An. superpictus* Grassi as a species complex including three genotypes X, Y, and Z and Djadid et al. (2009) introduced a

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in this species and it is not considered a vector in the country. Recently, Djadid et al. (2009) reported An. hyrcanus (Pallas) a potential vector of P. falciparum based on nested PCR in Guilan Province. West Nile and Sindbis viruses have been reported in Iran (Naficy and Saidi 1970). There is no information about their vectors in the country. The possibility of some mosquito-borne arboviral outbreaks like Japanese encephalitis and Rift Valley fever in the WHO Eastern Mediterranean Region, including Iran, is noteworthy (WHO 2004). There are some doubtful old records of Dengue fever in Iran (Foote and Cook 1959), however there is no formal recent record of this virus in the country. The mosquito-borne filariae: Dirofilaria (dirofilariasis) and Setaria (setariasis) (Spirurida: Onchocercidae) have been reported in Iran (Eslami 1997, Azari-Hamidian et al. 2007). Anopheles maculipennis and Culex theileri Theobald are known vectors of Setaria labiatopapillosa and Dirofilaria immitis respectively, in Ardebil Province, northwestern Iran (Azari-Hamidian et al. 2009).

In 1996, Qom Province was formally separated from Tehran Province. There is very little information about the mosquito fauna of Qom Province. Macan (1950) found An. multicolor larvae in "Darya-i-Namak" (Namak Lake, Salt Lake, or Qom Lake) (Fig. 1) and showed this species and An. superpictus around Qom in the distributional maps. In the document of the former Institute of Malariology and Parasitology (present National Institute of Health Research) (1953) An. multicolor was recorded around Qom. Saebi (1987) showed the occurrence of An. superpictus in Qom in the distributional map of the species. Recently, Farzinnia et al. (2010) reported An. claviger (Meigen) for the first time in the province. As the authors know there is no other documented information on the mosquitoes of Qom Province.

To study mosquito fauna and providing a primary checklist, an investigation was carried out on a collection of mosquitoes in Qom Province.

Materials and Methods

Study area

Qom Province is bounded by Tehran Province in the north, Isfahan Province in the south, Semnan Province in the east, and Markazi Province in the west with an area of approximately 11240 square km (0.68% total area of Iran). The center of the province, Qom City, is almost 880 m above sea level. The province with arid climate has about 150 mm annual rainfall and is located between $34^{\circ}09'-35^{\circ}11'$ N latitude and $50^{\circ}06'-51^{\circ}58'$ E longitude and formally includes one county (Qom) and five districts: Jafarabad, Kahak, Khalajestan, Markazi (Qom), and Salafchegan (Fig. 1).

Specimen and data collection

In the present faunistic and cross-sectional investigation, larvae were collected from different habitats using the standard dipping technique (350 ml dipper) in nine stations (localities) of four districts; Kahak District (Emamzadeh Esmail and Dastgerd), Khalajestan District (Ahmadabad, Agholak, Dastjerd, and Ghahan), Markazi District (Ghomrood), and Salafchegan District (Ghal-e-cham and Rahjerd) on 19 occasions during spring and summer 2008 and 2009. The larvae were preserved in lactophenol and the microscope slides of the preserved larvae were prepared using de Faure's medium. The third- and fourth-instar larvae were identified using the keys of Shahgudian (1960), Zaim and Cranston (1986), Harbach (1988), and Azari-Hamidian and Harbach (2009). The mosquito name abbreviations follow Reinert (2009).

Results

In total, 371 mosquito larvae were periodically collected from four districts of Qom Province during 2008–2009 and morphologically identified including 14 species representing four genera: An. claviger, An. marteri Senevet and Prunnelle, An. turkhudi Liston, An. superpictus, Culex arbieeni Salem, Cx. hortensis Ficalbi, Cx. mimeticus Noe, Cx. modestus Ficalbi, Cx. pipiens Linnaeus, Cx. territans Walker, Cx. theileri, Cu*liseta longiareolata* (Macquart), *Cs. subochrea* (Edwards), and *Ochlerotatus caspius* (Pallas) *s.l.* (Table 1). The association occasions of the mosquito larvae in the province are shown in Table 2. The association percentages of the mosquito larvae in Qom Province are shown in Table 3. All larvae were collected from natural habitats.

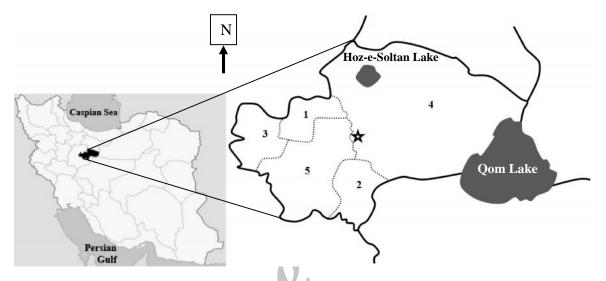


Fig. 1. Map of Iran, highlighting the position of Qom Province and its five districts: 1. Jafarabad, 2. Kahak, 3. Khalajestan, 4. Markazi, and 5. Salafchegan (* Qom City)

Table 1. The distribution and composition of the mosquito larvae in nine localities of Qom Province, Iran, Spring-
Summer 2008–2009

			V		Locality					_	
Species	Agholak	Ahmadabad	Dastgerd	Dastjerd	Emamzadeh Esmail	Ghahan	Ghal-e- cham	Ghomrood	Rahjerd	n	%
An. claviger	66	135	-	1	-	16	10	-	-	228	61.5
An. marteri	y _	-	-	-	-	-	-	-	2	2	0.5
An. superpictus	-	-	12	-	20	-	-	-	-	32	8.6
An. turkhudi	-	-	7	-	-	-	-	-	-	7	1.9
Cx. arbieeni	-	-	1	-	-	-	-	-	-	1	0.3
Cx. hortensis	3	2	1	-	-	3	-	-	-	9	2.4
Cx. mimeticus	-	-	9	-	1	1	-	-	-	11	3.0
Cx. modestus	-	-	-	-	-	-	-	6	-	6	1.6
Cx. pipiens	-	-	-	-	-	-	-	13	-	13	3.5
Cx. territans	7	3	-	-	-	-	-	-	-	10	2.7
Cx. theileri	-	1	-	-	4	-	-	1	-	6	1.6
Cs. longiareolata	-	34	-	-	-	5	-	-	2	41	11.1
Cs. subochrea	1	-	-	-	-	-	-	1	-	2	0.5
Oc. caspius s.l.	-	-	-	-	-	-	-	3	-	3	0.8
Total	77	175	30	1	25	25	10	24	4	371	100

Species	Total occasions	An. claviger	An. marteri	An. superpictus	An. turkhudi	Cx. arbieeni	Cx. hortensis	Cx. mimeticus	Cx. modestus	Cx. pipiens	Cx. territans	Cx. theileri	Cs. longiareolata	Cs. subochrea	Oc. caspius s.l.
An. claviger	14	-	-	-	-	-	4	1	-	-	2	1	3	1	-
An. marteri	1	-	-	-	-	-	-	-	-	-	-	-	1	-	-
An. superpictus	2	-	-	-	1	1	1	2	-	-	-	1	-	-	-
An. turkhudi	1	-	-	1	-	1	1	1	-	-	-			-	-
Cx. arbieeni	1	-	-	1	1	-	1	1	-	-	-	-	-	-	-
Cx. hortensis	5	4	-	1	1	1	-	2	-		1	1	2	1	-
Cx. mimeticus	3	1	-	2	1	1	2	-	-	-	\	1	1	-	-
Cx. modestus	1	-	-	-	-	-	-	-	- 1	1	-	1	-	1	1
Cx. pipiens	1	-	-	-	-	-	-	-	1	-	-	1	-	1	1
Cx. territans	2	2	-	-	-	-	1	-		-		-	1	1	-
Cx. theileri	3	1	-	1	-	-	1	1	1	1	- (-	-	1	1
Cs. longiareolata	4	3	1	-	-	-	2	1	-		1	-	-	-	-
Cs. subochrea	2	1	-	-	-	-	1	X	1	1	1	1	-	-	1
Oc. caspius s.l.	1	-	-	-	-	-	-	<u> </u>	1	1	-	1	-	1	-

Table 2. The association occasions of the mosquito larvae in Qom Province, Iran, Spring–Summer 2008–2009

Table 3. The association percentages of the mosquito larvae in Qom Province, Iran, Spring–Summer 2008–2009

Species association	Abundance n (%)
An. claviger	
Cx. hortensis, Cx. theileri	89 (39.0)
Alone	67 (29.4)
Cx. hortensis, Cx. territans, Cs. subochrea	59 (25.9)
Cx. territans, Cs. longiareolata	5 (2.2)
Cx. hortensis, Cx. mimeticus, Cs. longiareolata	5 (2.2)
Cx. hortensis, Cs. longiareolata	3 (1.3)
Total	228 (100)
An. marteri	
Cs. longiareolata	2 (100)
An. superpictus	
Cx. mimeticus, Cx. theileri	20 (62.5)
An. turkhudi, Cx. arbieeni, Cx. hortensis, Cx. mimeticus	12 (37.5)
Total	32 (100)
An. turkhudi	
An. superpictus, Cx. arbieeni, Cx. hortensis, Cx. mimeticus	7 (100)
Cx. arbieeni	
An. superpictus, An. turkhudi, Cx. hortensis, Cx. mimeticus	1 (100)
Cx. hortensis	
An. claviger, Cx. territans, Cs. subochrea	3 (33.4)
An. claviger, Cx. mimeticus, Cs. longiareolata	3 (33.3)
An. superpictus, An. turkhudi, Cx. arbieeni, Cx. mimeticus	1 (11.1)
An. claviger, Cs. longiareolata	1 (11.1)
An. claviger, Cx. theileri	1 (11.1)
Total	9 (100)

9 (81.8)
1 (9.1)
1 (9.1)
11 (100)
6 (100)
13 (100)
7 (70)
3 (30)
10 (100)
4 (66.6)
1 (16.7)
1 (16.7)
6 (100)
26 (63.4)
8 (19.5)
5 (12.2)
2 (4.9)
41 (100)
1 (100)
1 (100)
2 (100)
3 (100)

Discussion

In the present study, 371 mosquito larvae representing 14 species in four genera were collected from Qom Province. All species except for *An. claviger* and *An. superpictus* were collected for the first time in the province. *Anopheles multicolor*, which was recorded in Qom Province before (Macan 1950, Institute of Malariology and Parasitology 1953), was not found among the samples of this investigation.

Among the collected species, An. superpictus, Cx. hortensis, Cx. mimeticus, Cx. pipiens, Cx. theileri, Cs. longiareolata, Cs. subochrea, and Oc. caspius s.l. are widespread in Iran (Institute of Malariology and Parasitology 1953, Gaffary 1954, Saebi 1987, Zaim 1987, Abai et al. 2007). *Anopheles claviger* was found almost everywhere in Iran, including 21 provinces (out of total 31), except for the eastern and southeastern areas of the country. *Anopheles marteri* was collected from 15 provinces other than those of eastern and southeastern Iran. *Anopheles turkhudi* is found in 15 provinces in central plateau, western, southwestern, eastern, southeastern, and southern Iran. *Anopheles multicolor*, which was not found in this investigation, however recorded around Qom before, was recorded in 14 provinces in central plateau, northeastern, eastern, southeastern,

and southern Iran (Institute of Malariology and Parasitology 1953, Gaffary 1954, Saebi 1987). Culex arbieeni was recorded in 9 provinces before: former Khorasan, Loristan, Chaharmahal and Bakhtiari, Yazd, Kerman, Sistan and Baluchistan, Hormozgan, Fars, and Bushehr (Zaim 1987). Culex modestus was collected from 6 provinces: Isfahan, East Azerbaijan, West Azerbaijan, Hormozgan (Zaim 1987), Ardebil (Azari-Hamidian et al. 2009), and North Khorasan (Azari-Hamidian et al. 2011). Culex territans was found in 6 provinces: Guilan, Kurdistan, former Khorasan, Isfahan, Hormozgan (Zaim 1987), and Sistan and Baluchistan (Lotfi 1973). Among the species, only An. turkhudi and Cx. arbieeni are not found in northern Iran including northwestern areas (West Azerbaijan, East Azerbaijan, and Ardebil Provinces), the Caspian Sea littoral (Guilan, Mazandaran, and Golestan Provinces), and northeastern areas (North Khorasan Province and northern Razavi Khorasan Province) (Institute of Malariology and Parasitology 1953, Gaffary 1954, Saebi 1987, Zaim 1987).

Oshaghi et al. (2008) reported three genotypes named X, Y, and Z within An. superpictus in Iran based on the second Internal Transcribed Spacer (ITS2) and Cytochrome c Oxidase subunit I (COI) sequence data. Genotype X was found in all parts of the country, including Qom Province, except for the southeastern areas. Genotypes Y and Z were only found in southeastern Iran including Kahnooj of southern Kerman Province and Sistan and Baluchistan Province.

Only two larvae, one the third instar, of *Cs. subochrea* were found in the present investigation (Table 1). Two close and rare species *Cs. subochrea* and *Cs. annulata* (Schrank) are very similar in larval stage; however they are easily distinguishable as adult. There is little information about taxonomy and distribution of three close species *Cs. alaskaensis* (Ludlow), *Cs. annulata*, and

Cs. subochrea in Iran (Azari-Hamidian and Harbach 2009).

Among the species which were found in the present study, *An. superpictus* is malaria vector in Iran (Edrissian 2006). *Culex theileri* is known vector of the canine heart worm nematode in Ardebil Province (Azari-Hamidian et al. 2009). In addition, *Cx. pipiens*, *Cx. theileri*, and *Oc. caspius s.l.* are known as the potential or proven vectors of different human and domesticated animal pathogens in different areas of the world (Gutsevich et al. 1974, Harbach 1988). The ecology of medically important species and the unstudied areas of Qom Province need to be investigated extensively.

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References

- Abai MR, Azari-Hamidian S, Ladonni H, Hakimi M, Mashhadi-Esmail K, Sheikhzadeh K, Kousha A, Vatandoost H (2007) Fauna and checklist of mosquitoes (Diptera: Culicidae) of East Azerbaijan Province, northwestern Iran. Iran J Arthropod-Borne Dis. 1: 27–33.
- Azari-Hamidian S (2007) Checklist of Iranian mosquitoes (Diptera: Culicidae). J Vect Ecol. 32: 235–242.
- Azari-Hamidian S, Yaghoobi-Ershadi MR, Javadian E, Mobedi I, Abai MR (2007) Review of dirofilariasis in Iran. J Med Fac Guilan Univ Med Sci.

15(60): 102–113 [Persian with English abstract].

- Azari-Hamidian S, Harbach RE (2009) Keys to the adult females and fourth-instar larvae of the mosquitoes of Iran (Diptera: Culicidae). Zootaxa. 2078: 1–33.
- Azari-Hamidian S, Yaghoobi-Ershadi MR, Javadian E, Abai MR, Mobedi I, Linton YM, Harbach RE (2009) Distribution and ecology of mosquitoes in a focus of dirofilariasis in northwestern Iran, with the first finding of filarial larvae in naturally infected local mosquitoes. Med Vet Entomol. 23: 111– 121.
- Azari-Hamidian S, Abai MR, Arzamani K, Bakhshi H, Karami H, Ladonni H, Harbach RE (2011) Mosquitoes (Diptera: Culicidae) of North Khorasan Province, northeastern Iran and the zoogeographic affinities of the Iranian and Middle Asian mosquito fauna. J Entomol. 8: 204–217.
- Djadid ND, Jazayeri H, Gholizadeh S, Rad ShP, Zakeri S (2009) First record of a new member of *Anopheles* Hyrcanus Group from Iran: molecular identification, diagnosis, phylogeny, status of kdr resistance and *Plasmodium* infection. J Med Entomol. 46: 1084–1093.
- Edrissian GH (2006) Malaria in Iran: Past and present situation. Iran J Parasitol. 1: 1–14.
- Eshghy N (1977) *Anopheles multicolor* Cambouliu and its role in the transmission of malaria in Iran. J Ent Soc Iran. 4: 87–88 [Persian with English abstract].
- Eslami A (1997) Veterinary Helminthology. Vol. 3. Nematoda and Acanthocephala. Tehran University Publications, Tehran [Persian].
- Farzinnia B, Saghafipour A, Abai MR (2010) Malaria situation and anopheline mosquitoes in Qom Province, central Iran. Iran J Arthropod-Borne Dis. 4: 61–67.

- Foote RH, Cook DR (1959) Mosquitoes of Medical Importance. Agriculture Handbook No 152, Agricultural Research Service, US Department of Agriculture, Washington, DC.
- Gaffary EN (1954) Tentative distributional data on the anophelines of Iran. Annual Meeting of the American Mosquito Control association and the New Jersey Mosquito Extermination Association, Atlantic City, New Jersey, USA, p. 4.
- Gutsevich AV, Monchadskii AS, Shtakelberg AA (1974) Fauna of the USSR Diptera Volume III No 4 Mosquitoes Family Culicidae. Akad Nauk SSSR Zool Inst NS No 100, Leningrad [Russian, English translation by R Lavoott, ed by O Theodor, Israel Program for Scientific Translations, Jerusalem].
- Harbach RE (1988) The mosquitoes of the subgenus *Culex* in southwestern Asia and Egypt (Diptera: Culicidae). Contrib Am Ent Inst. 24(1): vi + 1–236.
- Harbach RE (2007 onwards) Mosquito Taxonomic Inventory. Available at: http://mosquito-taxonomic-inventory.info/ (accessed 20 Apr 2011).
- Institute of Malariology and Parasitology (1953) Distribution of *Anopheles* spp in Iran. Publication of Institute of Malariology and Parasitology No 225, Tehran [Persian].
- Lotfi MD (1973) Iranian species of genus *Culex* II Report of four species of larvae and 14 adult species. Bull Soc Pathol Exot. 66: 204–207.
- Macan TT (1950) *Anopheles* and Malaria in the Near East. Part III. The Anopheline Mosquitoes of Iraq and North Persia. London School of Hygiene and Tropical Medicine Research Memoir No 7, HK and Lewis Co Ltd, London.
- Naficy K, Saidi S (1970) Serological survey on viral antibodies in Iran. Trop Geogr Med. 2: 183–188.

- Oshaghi MA, Yaghobi-Ershadi MR, Shemshad Kh, Pedram M, Amani H (2008) The *Anopheles superpictus* complex: introduction of a new malaria vector complex in Iran. Bull Soc Pathol Exot. 101: 429–434.
- Reinert JF (2009) List of abbreviations for currently valid generic-level taxa in family Culicidae (Diptera). Eur Mosq Bull. 27: 68–76.
- Saebi ME (1987) Morphological study on anopheline larvae and their distribution in Iran. [PhD Dissertation]. School of Public Health, Tehran University of Medical Sciences, Tehran, Iran [Persian].

- Shahgudian ER (1960) A key to anophelines of Iran. Acta Med Iran. 3: 38–48.
- World Health Organization (2004) Integrated Vector Management. WHO Regional Officer for Eastern Mediterranean, Cairo.
- Zaim M, Cranston PS (1986) Checklist and keys to the Culicinae of Iran (Diptera: Culicidae). Mosq Syst. 18: 233–245.
- Zaim M (1987) The distribution and larval habitat characteristics of Iranian Culicinae. J Am Mosq Control Assoc. 3: 568–573.