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Culex mosquitoes (Diptera: Culicidae) recorded along the Nile River in central and northern Sudan, with a key for the identification of all species of the genus known to occur in the country

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Abstract

Despite the importance of *Culex* species as major vectors of Rift Valley fever virus, West Nile virus and the microfilariae that cause lymphatic filariasis, information on these mosquitoes in Sudan is limited to works published 65 years ago in the former Anglo-Egyptian Sudan, where some species were only recorded from areas of the territory now known as South Sudan. In this paper, we provide updated information on *Culex* mosquitoes collected indoors during surveillance studies conducted along the Nile River in central and northern areas of Sudan between 2012 and 2019. Of 3,411 female mosquitoes collected in Khartoum and northern states along the river, 2,560 (75%) were specimens of *Culex* belonging to 12 species: *Cx.* (*Culex*) *antennatus* (Becker, 1903), *Cx.* (*Cux.*) *laticinctus* Edwards, 1913, *Cx.* (*Cux.*) *neavei* Theobald, 1906, *Cx.* (*Cux.*) *pipiens* Linnaeus, 1758, *Cx.* (*Cux.*) *perexiguus* Theobald, 1903, *Cx.* (*Cux.*) *poicilipes* (Theobald, 1903), *Cx.* (*Cux.*) *quinquefasciatus* Say, 1823, *Cx.* (*Cux.*) *simpsoni* Theobald, 1905, *Cx.* (*Cux.*) *sinaiticus* Kirkpatrick, 1925, *Cx.* (*Cux.*) *theileri* Theobald, 1903, *Cx.* (*Cux.*) *tritaeniorhynchus* Giles, 1901 and *Cx.* (*Culiciomyia*) *macfiei* Edwards, 1923. This is the first record for *Cx. tritaeniorhynchus* and *Cx. macfiei* in central Sudan. The relative abundance of each species varied in different areas and seasons, but *Cx. antennatus* and *Cx. quinquefasciatus* were the most abundant indoor resting species. We provide an updated dichotomous key for the identification of the adults of *Culex* mosquitoes known to occur in the Republic of the Sudan.

Key words: bionomics, country records, *Lutzia*, mosquitoes, Rift Valley fever, surveys

Introduction

Culex Linnaeus, 1758 is the second largest genus of the family Culicidae (Diptera) with 777 species (Harbach 2020). The genus has a worldwide distribution and a number of species are implicated in the transmission of pathogens that cause several tropical diseases of humans, including the viruses (arboviruses) which cause Rift Valley fever and West Nile fever.

Many studies conducted in Africa provide strong evidence that many species are involved in the transmission of Rift Valley fever virus, for which members of the Pipiens Complex of the subgenus *Culex* are most frequently listed as vectors (Chevalier *et al*. 2010; Seufi & Galal 2010; Authie *et al*. 2013; Tantely *et al*. 2013). Other species of *Culex*, including *Cx. antennatus* (Becker, 1903), *Cx. bitaeniorhynchus* Giles, 1901, *Cx. neavei* Theobald, 1906, *Cx. perexiguus* Theobald, 1903, *Cx. poicilipes* (Theobald, 1903), *Cx. theileri* Theobald, 1903, *Cx. tritaeniorhynchus* Giles, 1901 and *Cx. univittatus* Theobald, 1901b, have been reported to be vectors of Rift Valley fever virus by vari-

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ous researchers (Hoogstraal *et al.* 1979; Diallo *et al.* 2000; Jupp *et al.* 2002; European Food Safety Authority 2005; Chevalier *et al*. 2010; Sang *et al.* 2010; Seufi & Galal 2010; Hanafi *et al.* 2011; Ratovonjato *et al.* 2011; Tantely *et al*. 2013).

Species of the Pipiens Complex are also the major vectors of West Nile virus (Al-Ali *et al.* 2008; Hamer *et al*. 2008), but *Cx*. *univittatus* and *Cx. tritaeniorhynchus* have also been implicated as vectors of the virus (Jupp 1996; Hubálek & Halouzka 1999; Tantely *et al*. 2013; Khan *et al*. 2017; Mavridis *et al*. 2018; Patsoula *et al*. 2020). In Sudan, Rift Valley fever outbreaks have been reported from the states of Gazeera, Kassala, Khartoum, River Nile, Sinnar and White Nile (Hassan *et al*. 2011). *Culex quinquefasciatus* Say, 1823 was incriminated as the main vector, as in other countries (Abdelgadir *et al*. 2010; Seufi & Galal 2010).

In addition to their role in the transmission of Rift Valley fever and West Nile viruses, *Culex* mosquitoes are important vectors of *Wuchereria bancrofti*, the microfilariae that cause lymphatic filariasis in urban and suburban areas in Africa. Studies indicate that *Cx. quinquefasciatus* is the most important vector (Derua *et al*. 2017).

Information on *Culex* mosquitoes in Sudan is limited to Edwards (1941) and Lewis (1956), whose records pertain to localities in the former Anglo-Egyptian Sudan. For this reason, the occurrence of some species in Sudan (o□cially the Republic of the Sudan) remains uncertain because they were recorded from localities in the present-day Republic of South Sudan. In this paper, we provide updated information on *Culex* mosquitoes collected resting in dwellings along the Nile River in the central and northern areas of Sudan, and provide a dichotomous key based on the species collected and the records of Edwards (1941) and Lewis (1956).

Material and methods

Study areas

Khartoum State. This state lies between latitudes 15° 10′ and 10° 30′ N and longitudes 32° 38′ and 34° 38′ E in the central area of Sudan. It includes the three towns of Khartoum, Khartoum North and Omdurman, and covers an area of 28,165 km². The area is semi-desert or impoverished savannah with little rainfall. Entomological surveys were carried out during 2012–2019 in the three aforementioned areas. Four sites were surveyed during 2012 and 2013 in the northern area of Omdurman and Khartoum North. The two sites in Omdurman were Haialarab (15° 38′ 47.45″ N, 32° 28′ 45.39″ E) and Abuseed (15° 35′ 51.01″ N, 32° 28′ 0.55″ E) and those in Khartoum North were Alkadro (15° 44′ 54.44″ N, 32° 33′ 43.81″ E) and Shendi Station (15° 37′ 30.16″ N, 32° 32′ 36.76″ E). Other surveys were carried out in 2013 and 2015 in the East Nile (agricultural, 15° 34′ 20.59″ N, 32° 41′ 11.41″ E) and Aldym (urban, 15° 34′ 54.94″ N, 32° 32′ 10.37″ E) sites, respectively. A final survey was conducted in 2019 at random sites in Khartoum (15° 30′ 7.60″ N, 32° 33′ 43.77″ E), Khartoum North and areas of Omdurman.

Nile State. This state is located between the 4th and 5th cataracts on the main Nile River and is accessible throughout the year because it lies on the railway line from Khartoum to Wadi Halfa. Entomological surveys were carried out in the Abu Hamad area (19° 3′ N, 33° 20′ E). The area is semi-desert with an annual rainfall of 10 mm. It is characterized by a cool minimum temperature (below 10 °C) during the main mosquito biting season in the winter. One site in the area, Algoz (19° 32′ 19.62″ N, 33° 18′ 59.20″ E), was surveyed during 2012 and 2013.

Northern State. This state is located between 16° 32′ N and 30° 32′ E, an area of about 348,765 km² situated approximately 350 km north of Khartoum. The state includes the Merowi area (Fig. 1) where two sites, Algorir (18° 18′ 19.60″ N, 31° 44′ 4.88″ E) and Nori (18° 33′ 41.86″ N, 31° 52′ 25.31″ E), were surveyed during 2012 and 2013. The area is desert to semi-desert with scant rainfall. In the summer months, May to September, the temperatures can rise to over 40 °C and sandstorms are common. The area is sparsely vegetated except along the banks of the Nile. Various varieties of palm trees flourish along the river and irrigation schemes allow quite intensive agriculture.

Mosquito surveillance and collection

Monthly collections of resting mosquitoes were made in 10 houses (one room in each house) over a period of one year in 2012 in the areas in the three states noted above. In 2013, a longitudinal survey was carried out during May–October (end of the hot dry-rainy season) in Khartoum North. This was achieved through two visits per week resulting in a total of 20 houses surveyed per month, except for August, when only four houses were inspected because of heavy floods in the area. In 2015, a longitudinal survey was carried out during January to July (end of cold dry-hot dry season) in Khartoum (two visits per week with a total of 20 houses/month). In 2019, cross-sectional sur-

veys were carried out from October 2018 to February 2019 (end of rainy cold-dry season) in 12 sites in Khartoum, Khartoum North and Omdurman (three visits/week with a total of 20 houses/month).

Collections were performed using the knock-down procedure according to the World Health Organization (1992). Knock down took place early in the morning after obtaining the consent of the occupants. Houses were selected based on the proximity to larval habitats and also to cover different types of buildings, especially in Khartoum State. Mosquitoes were preserved dry in labelled 1.5 ml Eppendorf tubes containing silica gel for subsequent morphological identification in the laboratory.

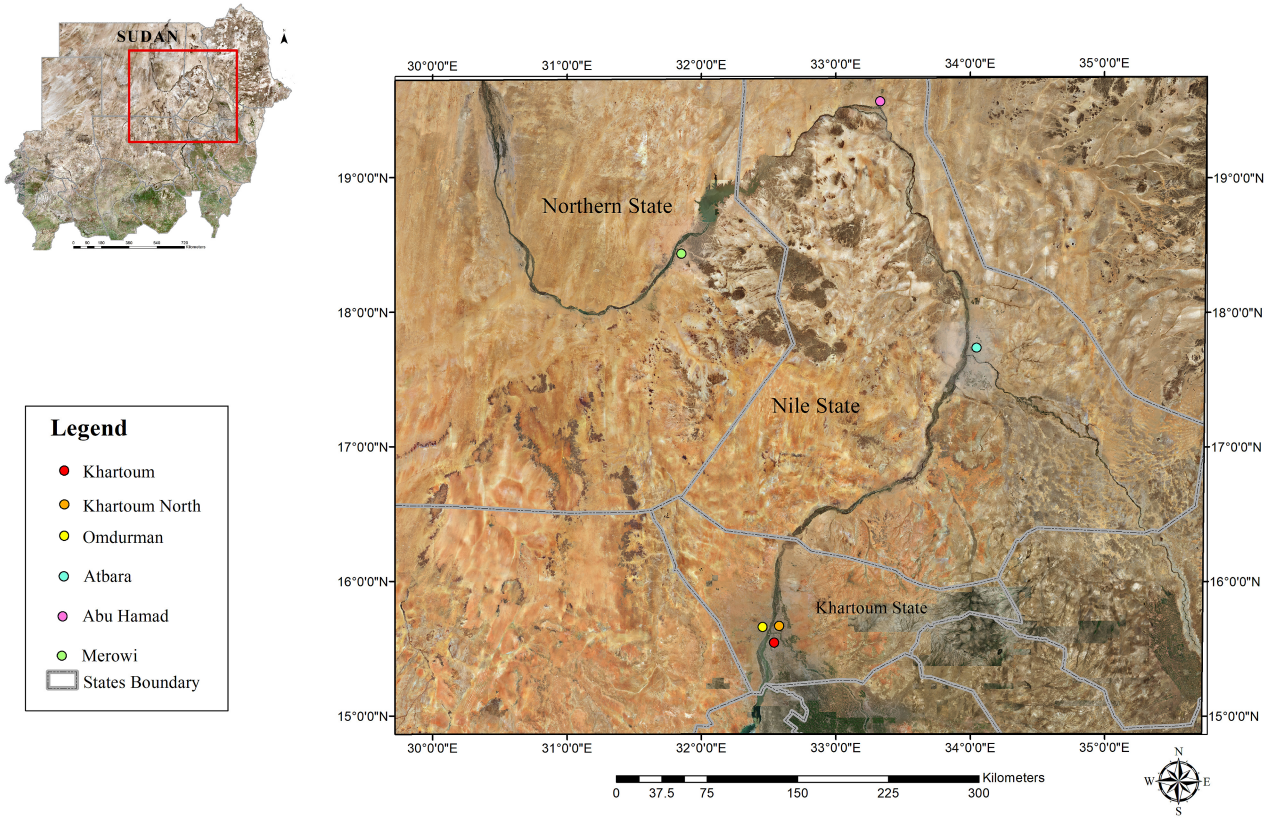

FIGURE 1. Landsat map showing the location of the study areas along the Nile River.

Identification of Culex *mosquitoes and preparation of keys*

Adult mosquitoes were identified to species using the keys of Edwards (1941) and Harbach (1988). Specimens were examined under a dissecting microscope with a magnification of 25x. The dichotomous key below was prepared to identify the species of *Culex* known to occur in central and northern Sudan and other areas of the country. The key includes the species collected during the study and those recorded by Edwards (1941) and Lewis (1956) from localities in the study area. The morphological terminology of Harbach & Knight (1980, 1982), revised and updated in the Anatomical Glossary of the Mosquito Taxonomic Inventory (http://mosquito-taxonomic-inventory.info/), is used in the keys. The generally accepted two-letter abbreviations for the genera *Culex* and *Lutzia*, i.e. *Cx.* and *Lt.*, and the three-letter abbreviations recommended by Reinert (2009) are used for the subgenera of *Culex*, including: *Cui.* = *Culiciomyia* Theobald, 1907; *Cux.* = subgenus *Culex*; *Eum.* = *Eumelanomyia* Theobald, 1909; *Ocu.* = *Oculeomyia* Theobald, 1907.

Results

A total of 3,411 indoor-resting female mosquitoes were collected during the study, among which 2,560 (75%) were identified as members of 12 species of *Culex* representing two subgenera: *Cx.* (*Culex*) *antennatus*, *Cx.* (*Cux.*) *laticinctus* Edwards, 1913, *Cx.* (*Cux.*) *neavei*, *Cx.* (*Cux.*) *pipiens* Linnaeus, 1758, *Cx.* (*Cux.*) *perexiguus*, *Cx.* (*Cux.*) *poicilipes*, *Cx.* (*Cux.*) *quinquefasciatus*, *Cx.* (*Cux.*) *simpsoni* Theobald, 1905, *Cx.* (*Cux.*) *sinaiticus* Kirkpatrick,

1925, *Cx.* (*Cux*.) *theileri*, *Cx.* (*Cux*.) *tritaeniorhynchus* and *Cx.* (*Culiciomyia*) *macfiei* Edwards, 1923. *Culex tritaeniorhynchus* and *Cx. macfiei* are recorded from central Sudan for the first time. The relative abundance of each of the species varied between different areas and seasons.

The first survey was carried out in 2012 in seven areas along the Nile River. Of 1,914 female mosquitoes collected, 1,691 (88.3%) were identified as members of the following nine *Culex* species: *Cx*. *antennatus* (1,143, 67.6%), *Cx. macfiei* (3, 0.2%), *Cx*. *pipiens* (53, 3.1%), *Cx*. *quinquefasciatus* (391, 23.1%), *Cx*. *simpsoni* (38, 2.2%), *Cx*. *sinaiticus* (13, 0.8%), *Cx*. *theileri* (1, 0.06%), *Cx*. *tritaeniorhynchus* (1, 0.06%) and *Cx*. *univittatus* (48, 2.8%).

Consecutive surveys conducted in agricultural areas of Khartoum State during the rainy season in 2013 showed that out of 630 indoor-resting mosquitoes, *Culex* species comprised the lowest percentage (96, 15.2%). The most numerous *Culex s*pecies was *Cx. antennatus* (47, 49%), followed by *Cx. quinquefasciatus* (19, 19%), *Cx. sinaiticus* (12, 12.5%), *Cx. univittatus* (8, 8.3%), *Cx. laticinctus* (3, 3.1%), *Cx. poicilipes* (3, 3.1%), *Cx. simpsoni* (3, 3.1%) and *Cx. neavei* (1, 1.04%).

The collections made in 2015 and 2019 in urban areas of Khartoum State included females of three *Culex* species each year. Those collected during the hot-dry season in 2015 included *Cx. simpsoni* (154, 47.5%), *Cx. quinquefasciatus* (7, 2.2%) and *Cx. sinaiticus* (163, 50.3%); those collected during the cold-dry season in 2019 included *Cx. antennatus* (81, 18%), *Cx. quinquefasciatus* (249, 55.5%) and *Cx. univittatus* (119, 26.5%).

The numbers and percentages of the 12 species of *Culex* collected resting indoors along the Nile River in Khartoum State during 2012 to 2019 are given in Table 1.

Discussion

All *Culex* species captured during the study were recorded by Lewis (1956), with the exception of *Cx*. *macfiei* and *Cx. tritaeniorhynchus*, which are recorded here for the first time in northern Sudan. Lewis mentioned that specimens of *Cx. pipiens* were collected in lowland areas at several places between Omdurman in Khartoum State and Wadi Halfa in Northern State. He believed that the species was "spread by steamers up the White Nile, and by train to several other lowland places near the Nile and to the coastal area". However, molecular identification or dissection of male genitalia is needed to confirm the identity of females and larvae identified as *Cx. pipiens*. *Culex pipiens* and *Cx*. *quinquefasciatus* are both important vectors of Rift Valley fever and West Nile viruses. These species are principally ornithophilic (Gad *et al*. 1999; Simpson *et al*. 2009; Montgomery *et al*. 2011; Gomes *et al*. 2013); therefore, we expect that the number of specimens residing outdoors is much greater than the number of specimens collected indoors.

Of the 12 *Culex* species collected resting indoors along the Nile in northern Sudan, eight have been implicated as vectors of Rift Valley fever virus, i.e. *Cx. antennatus*, *Cx. neavei*, *Cx. pipiens*, *Cx. poicilipes*, *Cx. quinquefasciatus*, *Cx. theileri*, *Cx. tritaeniorhynchus* and *Cx. univittatus*. Additionally, some of these species have been implicated as vectors of West Nile virus, i.e. *Cx. pipiens*, *Cx. quinquefasciatus, Cx. tritaeniorhynchus* and *Cx. univittatus*. These findings point to the urgent need for vector control that targets species of *Culex* in addition to species of *Aedes* Meigen, 1818 and *Anopheles* Meigen, 1818.

The present study found that *Cx. antennatus* is the dominant indoor-resting species of *Culex*. Lewis (1956) reported that *Cx. antennatus* is widely distributed in Sudan, and females mainly feed on humans. Becker *et al.* (2010) stated that *Cx. antennatus* is a voracious biter and an important vector—the females primarily feed on livestock and humans both indoors and outdoors and are a major nuisance in suburban areas in Africa. *Culex antennatus* has been recorded as a vector of Rift Valley fever and West Nile viruses (Seufi & Galal 2010; Hanafi *et al.* 2011). During an outbreak of Rift Valley fever in the Nile Delta of Egypt, Hanafi *et al*. (2011) recorded *Cx. antennatus* as the dominant species. Records from Egypt show the presence of *Cx. antennatus*, *Cx. pipiens* and *Cx. univittatus* in indoor-resting collections (Beier *et al*. 1986). The continuous presence of *Cx. antennatus* in high numbers could be attributed to the availability of suitable larval habitats, including stream pools, springs, ponds, swamps, ditches, seepages and animal footprints that usually contain emergent vegetation (Becker *et al*. 2010). In Khartoum State, Abu Groon (1988) and Elmalih & Hassan (2018) found that the dominant *Culex* species was *Cx. quinquefasciatus*, which might be explained by the use of different collection methods. The higher percentages of *Culex* mosquitoes collected in the Omdurman area might be explained by unplanned urbanization where polluted aquatic sites with high organic content, including sewage, are common around or within houses (World Health Organization 2013). This can also explain the endophilic biting activity reported by Brown & Pal (1971).

TABLE 1. Total number and percentage of indoor resting female mosquitoes of the 12 species of *Culex* captured along the Nile River during the study. Coordinates of the specific collection sites are given in the Material and methods.

Year	**2012**							2013	2015	2019
State	**Khartoum State**				**Northern State**		**Nile State**	**Khartoum State**		
Area	Omdurman		Khartoum North		Merowi		Abu hamad	**Khartoum North**	**Khartoum, Khartoum North and Omdurman**	
Site / Species	**Haialarab**	**Abuseed**	**Shendi Station**	**Alkadro**	**Algorir**	**Nori**	**Algoz**	**East Nile**	**Aldym**	**Twelve different Sites**
Cx. antennatus	489	141	178	93	25	38	179	47	0	81
	(74.1)	(64.7)	(65.7)	(54.7)	(61)	(73.1)	(64.2)	(49)	(0.0)	(18)
Cx. laticinctus	0	0	0	0	0	0	0	3	0	0
	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(3.1)	(0.0)	(0.0)
Cx. macfiei	3	0	0	0	0	0	0	0	0	0
	(0.5)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)
Cx. neavei	0	0	0	0	0	0	0	1	0	0
	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(1.04)	(0.0)	(0.0)
Cx. pipiens	13	4	11	16	1	0	8	0	0	0
	(2.0)	(1.8)	(4.0)	(9.4)	(2.4)	(0.0)	(2.9)	(0.0)	(0.0)	(0.0)
Cx. poicilipes	0	0	0	0	0	0	0	3	0	0
	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(3.1)	(0.0)	(0.0)
Cx. quinquefasciatus	127	38	64	49	10	11	92	18	7	249
	(19.2)	(17.4)	(23.6)	(28.8)	(24.4)	(1.1)	(32.9)	(19)	(2.2)	(55.5)
Cx. simpsoni	14	11	5	5	3	0	0	3	154	0
	(2.1)	(5.0)	(1.8)	(2.9)	(7.3)	(0.0)	(0.0)	(3.1)	(47.5)	(0.0)
Cx. sinaiticus	3	5	2	1	1	1	0	12	163	0
	(0.5)	(2.3)	(0.7)	(0.6)	(2.4)	(1.9)	(0.0)	(12.5)	(50.3)	(0.0)
Cx. theileri	0	0	1	0	0	0	0	0	0	0
	(0.0)	(0.0)	(0.4)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)
Cx. tritaeniorhynchus	0	1	0	0	0	0	0	0	0	0
	(0.0)	(0.5)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)	(0.0)
Cx. univittatus	11	18	10	6	1	2	0	8	0	119
	(1.6)	(8.3)	(3.7)	(3.5)	(2.4)	(3.8)	(0.0)	(8.3)	(0.0)	(26.5)
Total (%)	660	218	271	170	41	52	**279**	**96**	**324**	**449**
	(100)	**(100)**	(99.9)	(99.9)	(99.9)	**(99.9)**	**(100)**	**(99.8)**	**(100)**	**(100)**

Sixteen genera of mosquitoes are recognized in the Republic of the Sudan, and keys to those genera are included in Mohamed *et al*. (2017). Based on the findings of the present study and the occurrence records of Edwards (1941) and Lewis (1956), 26 species of *Culex* are known to occur in Sudan, with 22 known to occur in the northern region of the country where this study was conducted (Table 2). A dichotomous key is provided below for the identification of all 26 species recorded in the country. Species of *Culex* included in the key that are not mentioned in the text above include *Cx.* (*Cui.*) *cinereus* Theobald, 1901b, *Cx.* (*Cui.*) *nebulosus* Theobald, 1901c, *Cx.* (*Cux.*) *argenteopunctatus* subsp. *kingii* (Theobald, 1913), *Cx.* (*Cux.*) *decens* Theobald, 1901c, *Cx.* (*Cux*.) *duttoni* Theobald, 1901c, *Cx.* (*Cux.*) *grahamii* Theobald, 1910, *Cx.* (*Cux.*) *perfuscus* Edwards, 1914, *Cx*. (*Cux*.) *sitiens* Wiedemann, 1828, *Cx.* (*Cux.*) *weschei* Edwards, 1935, *Cx.* (*Eum.*) *inconspicuosus* (Theobald, 1908), *Cx.* (*Eum.*) *kingianus* Edwards, 1922, *Cx.* (*Eum.*) *simpliciforceps* Edwards, 1941 and *Cx.* (*Ocu.*) *annulioris* Theobald, 1901a. The single species of *Lutzia* Theobald, 1903 that occurs in the country, i.e. *Lt. tigripes* (de Grandpré & de Charmoy, 1901), is included in the key because *Lutzia* was historically classified as a subgenus of *Culex* and its current generic status established by Tanaka (2003) is uncertain (Kitching *et al*. 2015; Harbach *et al*. 2017); however, Sun *et al*. (2019) provided evidence based on complete mitochondrial genomes that supports the generic status of *Lutzia*.

TABLE 2. Species of *Culex* recorded in central and northern Sudan.

Species	Edwards (1941)	Lewis (1956)	Present authors
Cx. antennatus (Becker)	+	+	+
Cx. annulioris Theobald	-	+	-
Cx. argenteopunctatus subsp. *kingii* (Theobald)	+	+	-
Cx. bitaeniorhynchus Giles	-	+	-
Cx. decens Theobald	+	+	-
Cx. duttoni Theobald	-	+	-
Cx. grahamii Theobald	-	+	-
Cx. laticinctus Edwards	+	+	+
Cx. macfiei Edwards	-	-	+
Cx. neavei Theobald	+	+	+
Cx. nebulosus Theobald	+	+	-
Cx. perfuscus Edwards	+	+	-
Cx. pipiens Linnaeus	+	+	+
Cx. poicilipes (Theobald)	+	+	+
Cx. quinquefasciatus Say	+	+	+
Cx. simpsoni Theobald	+	+	+
Cx. sinaiticus Kirkpatrick	+	+	+
Cx. sitiens Wiedemann	+	+	-
Cx. theileri Theobald	-	+	+
Cx. tritaeniorhynchus Giles	-	-	+
Cx. univittatus Theobald	+	+	+
Cx. weschei Edwards	-	+	-

Key for the adults of *Culex* and *Lutzia* species known to occur in northern Sudan

1 Normally 4 or more lower mesepimeral setae; fore- and midfemora and -tibiae each with an anterior row of small pale spots . . . . . *Lt. tigripes*

- A single lower mesepimeral seta (exceptionally 2 or 3) or none; femora and tibiae without rows of pale spots (except *Cx. poicilipes*) 2

2(1) Acrostichal setae absent . . . . . *Cx.* (*Cui.*) *macfiei*

- Acrostichal setae present, may be quite small 3

3(2) No lower mesepimeral seta; proboscis and tarsi with pale rings 4

- 1–3 lower mesepimeral setae 8

4(3) Femora and tibiae with rows of small pale spots anteriorly . . . . . *Cx.* (*Ocu.*) *poicilipes*
- Femora and tibiae without pale spots 5
5(4) Abdominal terga with apical pale bands; wing with numerous scattered pale scales; proboscis with a pair of dorsolateral pale spots before the labella . . . . . *Cx.* (*Ocu.*) *bitaeniorhynchus*
- Abdominal terga without apical pale bands; wing with few or no pale scales; proboscis dark at the apex (but the labella may be pale) 6
6(5) Thorax brownish or scutum with many pale scales; scutum usually with anterior 0.5 mainly pale-scaled; wing without pale scales at the apex, length 4.0–6.5 mm; abdominal terga with basomedian and apicolateral triangular patches of pale scales . . . . . *Cx.* (*Ocu.*) *annulioris*
- Scutal scales form an indefinite mottled pattern, or scales all dark; wing length 2.0–4.0 mm 7
7(6) Femora with numerous scattered pale scales anteriorly; cell R_2 of wing short, its base distal to the base of cell M_1; pale scales of the proboscis confined to a distinct median ring . . . . . *Cx.* (*Cux.*) *sitiens*
- Femora without or with a few scattered pale scales; cell R_2 longer, its base at least slightly proximal to the base of M_1; pale scaling of the proboscis in a ring with a proximal extension on the ventral surface . . . . . *Cx.* (*Cux.*) *tritaeniorhynchus*
8(3) Abdominal terga with basal pale bands (except *Cx. argenteopunctatus* subsp. *kingii* and some *Cx. neavei*) or lateral pale spots; vertex and ocular line of the head with narrow decumbent scales 9
- Abdominal terga completely dark-scaled or with apical pale scaling 24
9(8) Tarsi, especially hindtarsi, with narrow pale rings; prealar scales absent; midtibia with a pale anterior stripe; postspiracular scales usually present . . . . . *Cx.* (*Cux.*) *duttoni*
- Tarsi entirely dark-scaled; midtibia with or without an anterior pale stripe; postspiracular scales usually absent 10
10(9) Femora of fore- and midlegs and tibiae of all legs with anterior pale stripes running the whole length; hindtibia without a dorsal pale spot at the apex; prealar and upper and lower mesokatepisternal scale-patches contiguous, forming a single patch . . . . . *Cx.* (*Cux.*) *theileri*
- Femora and tibiae either without anterior stripes or if stripes are present on any of them then hindtibia with a dorsal pale spot at the apex; prealar and upper and lower mesokatepisternal scale-patches not continuous, in 3 separate patches 11
11(10) Scutum with 4 spots of silvery-white scales; small dark species, abdominal terga without pale bands . . . . . *Cx.* (*Cux.*) *argenteopunctatus* subsp. *kingii*
- Scutum without silvery-white spots 12
12(11) Postspiracular and prealar scales present; hindfemur with anterior surface mainly white-scaled, at most distal 0.2 dark-scaled 13
- Postspiracular scales absent; prealar scales normally present 17
13(12) Hindtibia with distinct or indistinct anterior and posterior pale stripes; wing with or without a short line of pale scales at the base of the costa 14
- Hindtibia without an anterior pale stripe, apex with a conspicuous white spot; wing entirely dark-scaled 16
14(13) Anterior surface of midfemur with distinct or indistinct anterior pale stripe; hindtibia with distinct anterior and posterior pale stripes on proximal 0.8, with distinct apical pale spot; costa of wing with a short line of pale scales at the base; patch of scales covers more or less of the dorsal 0.5 of the postspiracular area; pale bands of abdominal terga with normal basal pale bands 15
- Anterior surface of midfemur normally without an anterior pale stripe, weakly indicated when present; hindtibia with rather indistinct anterior and posterior pale stripes ending before the base, with rather an indistinct apical pale spot; wing entirely dark-scaled, without pale scales at the base of the costa; scales of postspiracular area tend to occur in a small patch near the spiracle; pale bands of the abdominal terga reduced or absent . . . . . *Cx.* (*Cux.*) *neavei*
15(14) Proboscis with pale scales on middle of ventral surface; midfemur with a complete distinct or indistinct anterior pale stripe; anterior and posterior pale stripes of hindtibia separated ventrally by a complete dark stripe; wing vein 2A (posterior to vein 1A) of female usually with a line of scales . . . . . *Cx.* (*Cux.*) *univittatus*
- Proboscis pale-scaled ventrally except at base, weakly on distal 0.25; midfemur with or without an incomplete faint or distinct anterior pale stripe; anterior and posterior pale stripe of hindtibia partly separated on proximal 0.5 or less by a weak ventral dark stripe; wing vein 2A of female occasionally with a few scales . . . . . *Cx.* (*Cux.*) *perexiguus*
16(13) Hindfemur with complete or nearly complete anterodorsal dark stripe; females: abdominal sterna with apical dark bands; forecoxa with some dark scales; males: abdominal sterna mainly dark-scaled, with basolateral pale spots, maxillary palpus normal . . . . . *Cx.* (*Cux.*) *simpsoni*
- Hindfemur with an anterodorsal dark stripe on distal 0.5 or less; females: abdominal sterna usually entirely pale-scaled; scales of forecoxa usually all pale; males: abdominal sterna mainly pale-scaled, posterior sterna usually with dark scales posteriorly, maxillary palpus sparsely setose . . . . . *Cx.* (*Cux.*) *sinaiticus*
17(12) Abdominal terga with basal pale bands 18
- Abdominal terga, at least the first few, without pale bands; mesepimeron with a distinct scale-patch in the middle 20
18(17) Thorax pale; proboscis entirely dark-scaled; abdominal sterna pale-scaled; 2–4 lower mesepimeral setae frequently present; hindtibia with conspicuous apical pale spot . . . . . *Cx.* (*Cux.*) *laticinctus*
- Thorax darker; proboscis pale beneath in the middle; only 1 lower mesepimeral seta; hindtibia with inconspicuous apical pale spot 19
19(18) Scutal scales golden brown with reddish tint; wing of female with cell M_2 more than 3 times as long as its stem (vein R_{2+3}), subcosta normally intersects the costa at or beyond the furcation of R_{2+3}; basal bands of abdominal terga yellowish, usually same color as the sternal scales; tergal bands of female distinctly darker than basolateral white spots . . . . . *Cx.* (*Cux.*) *pipiens*
- Scutal scales more or less buff-colored; wing of female with cell R_2 2.8–3.3 times as long as vein R_{2+3}, subcosta normally in-

tersects the costa before the furcation of R_{2+3}; basal bands of abdominal terga nearly white, usually slightly paler than sternal scaling, tergal bands of female slightly if at all darker than basolateral white spots . . . . . *Cx.* (*Cux.*) *quinquefasciatus*

20(17) Antenna of female normal, each flagellomere with 4–6 long setae; knob of halter usually dark-scaled. 21

- Antenna of female sub-verticillate; first few flagellomeres each with 10–20 long setae; halter entirely yellow-scaled. 23

21(20) Small brown species; abdominal terga without basal pale bands, terga VI and VII with lateral pale stripes; sterna pale-scaled . . . . . *Cx.* (*Cux.*) *antennatus*

- Larger species; abdominal terga with or without narrow basal pale bands, with basolateral pale patches; sterna with apical dark bands 22

22(21) Erect scales of head all or nearly all dark; scutum with evenly dispersed uniform reddish-brown scales, anterior and lateral margins and prescutellar area with pale scales; hindfemur with 0.2 or less of anterior surface dark-scaled; hindtibia with a pale spot at the apex . . . . . *Cx.* (*Cux.*) *decens*

- Erect scales of head all dark; scales on margins of scutum and prescutellar area creamy-white; hindfemur with anterior dark area at apex longer than broad; hindtibia entirely dark-scaled . . . . . *Cx.* (*Cux.*) *perfuscus*

23(20) Proboscis dark-scaled; 2 lower mesepimeral setae; hindtibia with a spot of pale scales at the apex; abdominal sterna with dark apical bands . . . . . *Cx.* (*Cux.*) *grahamii*

- Proboscis pale beneath; usually only 1 lower mesepimeral seta; hindtibia entirely dark-scaled; abdominal sterna entirely pale-scaled or with only a few dark scales on the apical margins . . . . . *Cx.* (*Cux.*) *weschei*

24(8) Vertex of head with narrow whitish decumbent scales; mesepimeron with very few or no scales . . . . . *Cx.* (*Eum.*) *kingianus*

- Vertex with some moderately broad scales, at least on the ocular line 25

25(24) Only ocular line of head with broad scales, the scales white; abdominal terga with apical lateral pale spots or all dark; thoracic pleura gray-dusted 26

- Vertex of head usually with more numerous broad scales, and these usually dark 27

26(25) Small species; length of wing 3–4 mm; mesepimeron usually without distinct scale-patch; gonocoxite of male genitalia with few short setae . . . . . *Cx.* (*Cui.*) *nebulosus*

- Larger species; length of wing 4–5 mm; mesepimeron usually with a large patch of scales; gonocoxite of male genitalia with dense patch of soft pale setae . . . . . *Cx.* (*Cui.*) *cinereus*

27(25) Dorsum of head almost entirely clothed in broad decumbent scales; legs mainly dark-scaled; hindfemur only indistinctly pale-scaled ventrally . . . . . *Cx.* (*Eum.*) *inconspicuosus*

- Dorsum of head with a fairly broad band of decumbent scales in front adjoining the eyes; hindfemur more extensively pale-scaled, ventral surface distinctly paler . . . . . *Cx.* (*Eum.*) *simpliciforceps*

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