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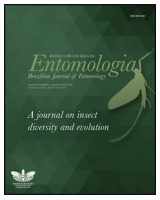

A survey of *Stomoxys* Geoffroy, 1762 (Diptera: Muscidae) in eight administrative regions of Cameroon

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ABSTRACT

Stomoxys Geoffroy, 1762 are major livestock pests in the tropics and are common in diverse habitats. This study aims to conduct a survey on the *Stomoxys* fauna of Cameroon. From 2015 to 2017, entomological studies using standard traps (n=204) were conducted in eight administrative regions found in five agro-ecological zones (AEZs) of Cameroon with 606 trap-points over 22,032 traps days. A total of 77,804 *Stomoxys* specimens were collected, with eight taxa consisting of six species (*S. calcitrans* (Linnaeus, 1758), *S. omega* Newstead, Dutton & Todd, 1907, *S. xanthomelas* Roubaud, 1937, *S. inornatus* Grunberg, 1906, *S. transvittatus* Villeneuve, 1916, and *S. sitiens* (Rondani, 1873)) and two subspecies (*Stomoxys niger niger* Macquart, 1851 and *S. niger bilineatus* Grunberg, 1906) identified. Among all the recorded taxa, *S. calcitrans* and *S. n. niger* were present in five and seven of the eight regions respectively, but *S. sitiens* was rare and only found in the North. Furthermore, the highest species number (seven out of eight) was recorded in the Guinee savanna of the Adamawa region. The highest apparent density range of 101 to 200 *Stomoxys*/ trap/ day (s/t/d) was recorded in the Sudan savanna AEZ of the Far North region. *Stomoxys* occurred in all the AEZs in eight regions, some of which are major cattle rearing regions. This represents risk for the mechanically transmission of dangerous pathogens in those regions.

Introduction

Stomoxys flies are blood sucking arthropods, dipterous insects belonging to the family Muscidae, subfamily Stomoxyinae, which includes 10 different genera (Zumpt, 1973). Eighteen species are known in the world to belong to the genus *Stomoxys,* and 17 of them have a tropical distribution (Zumpt, 1973). Large numbers of *Stomoxys* attack livestock, wild animals, and occasionally humans, as both males and females are bloodsuckers (Moon, 2002). In the Far North region of Cameroon, *Stomoxys* flies host preferences include cattle, horses, donkeys, humans, and poultry (Mamoudou et al., 2016). An investigation regarding the preferred breeding substrates for *S. calcitrans* showed a high propensity for egg-laying in vertebrate-herbivore dung, caused by signature odours emanating from the dung (Baleba et al., 2019). However, *Stomoxys* have a broad range of habitats and are continuously adapting to exploit new breeding substrates in forest, livestock, and human settlement areas (Mavoungou et al., 2017).

Stomoxys strong association with livestock, mainly to acquire a bloodmeal, creates severe adverse effects because high densities of flies (≥14 *Stomoxys* per animal per day) often leads to significant reduction in livestock weight gain and milk yield (Miller et al., 1973; Campbell et al., 2001; Lendzele et al., 2019). For pastured cattle in the USA, economic losses caused by *Stomoxys* have been estimated at $2.21 billion annually (Taylor et al., 2012). Due to these adverse direct effects on livestock, farmers in the Northern regions of Cameroon have constructed livestock houses with mosquito net windows to protect their animals during the stable fly's outbreak seasons (Sevidzem et al., 2019d). Apart from the direct effects caused by *Stomoxys* on livestock such as nuisance and blood sucking

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behaviours, *Stomoxys* can indirectly serve as mechanical vectors of several pathogens (bacteria, viruses, and protozoa) (Foil and Gorham, 2003; Baldacchino et al., 2013). In the Central African sub-region where Cameroon is located, *Stomoxys* flies have been reported (Sevidzem et al., 2016; Mavoungou et al., 2017), with some cross-sectional studies in the country identifying a few species (Sevidzem et al., 2016; Mounioko et al., 2018; Hiol et al., 2019; Mamoudou et al., 2020; Sevidzem et al., 2023). In neighbouring Nigeria two taxa comprising of one subspecies (*S. niger niger*) and one species (*S. calcitrans*) have been reported (Odeniran et al., 2019), while seven taxa comprising of five species (*S. calcitrans*, *S. xanthomelas*, *S. inornatus*, *S. transvittatus*, and *S. omega*) and two subspecies (*S. niger niger* and *S. niger bilineatus*) have been reported in neighbouring Gabon (Bitome-Essono et al., 2015; Mavoungou et al., 2017; Mounioko et al., 2018; Sevidzem et al., 2019b). The previous studies were only conducted in one of the different administrative regions and Agro-ecological Zones (AEZs) (Sevidzem et al., 2021) of Cameroon, but the present study will cover all of them. To the best of our knowledge no exhaustive study has been conducted to determine the abundance and distribution of *Stomoxys* species in Africa. The present study aimed to conduct a survey on the *Stomoxys* fauna of Cameroon.

Materials and methods

Study area

This study was conducted in eight different regions (Adamawa, North, Far North, East, South, Center, Littoral, North West) (Fig. 1a) located in the five main AEZs of Cameroon (Fig. 1b). The geographical coordinates and climatic characteristics of the different AEZs include, (i) the Sudan savanna, which covers the Far North (11°00′N, 14°30′E) and North (7°77796'N, 14°929'E) regions at elevations from 304 to 545 m above sea level (*a.s.l.*), respectively. The climate is considered Sudano-Sahelian with two seasons (rainy and dry), with an average monthly temperature of 28°C, and 400 to 1200 mm/year of rainfall; (ii) The Guinean savanna, which covers the Adamawa Plateau (7°00334'N, 13°01'E) at an elevation of 1000 m *a.s.l.* with a Sudanese climate type and average monthly temperature of 20 to 26°C, and rainfall of 1500 mm/year; (iii) The Highland plateau, covering the North West region (5°92523'N, 10°009'E) at an elevation of 900 m *a.s.l.* This region has a cold climate with mean monthly temperature of 19°C and annual rainfall of 1500 to 2000 mm/year; (iv) The Rainforest, covering the Littoral region (3°23333' N, 9°567' E) at an elevation of 35 m *a.s.l.* This region is a variable equatorial climate that is humid and hot, with average monthly temperatures between 22 to 29°C. The rainfall reaches 2500-4000 mm/year, and; (v) The Mosaic forest, which covers the East region (6°23333'N, 13°25'E) at an elevation of 890 m *a.s.l.* It has a Guinean climate (hot and humid) and rainfall of 1500 to 2000 mm/year (Sevidzem et al., 2021). Cameroon is a major cattle rearing region and supplier of livestock products to neighbouring countries in the central African sub-region (Mamoudou and Sevidzem, 2017). The major livestock rearing regions of Cameroon are the Far North, Adamawa, North and North West (Mamoudou and Sevidzem, 2017).

The livestock population in Cameroon includes: 31 million poultry, 6 million cattle, 7 million small ruminants, one million pigs, 150000 donkeys and 15000 horses (FAO, 2015). The distribution of cattle throughout the national territory is as such: 37.5% in the Far North, 33.9% in Adamawa, 11.6% in the North, 8% in the North West, 6.3% in the East and 2.7% in the West (Moumini, 2011).

Entomological studies

Stomoxys trapping was conducted in eight administrative regions from 2015 to 2017. Twenty eight sites were studied in the five main AEZs of Cameroon; characteristics of each trap point, trap model, trap numbers, time interval and trapping period are summarized in Table 1. Four different blue-black cloth trap types such as Nzi, Vavoua, Biconical, and MVT (Sevidzem et al., 2019a) were used. Due to the variation in topography, limited number of traps, and personnel for monitoring, the number of traps and trap types varied between sites. The localisation of trap-points was conducted using a Global Positioning System (GPS) handset (GPS eTrex®; Garmin (Europe) Ltd, Southampton, UK).

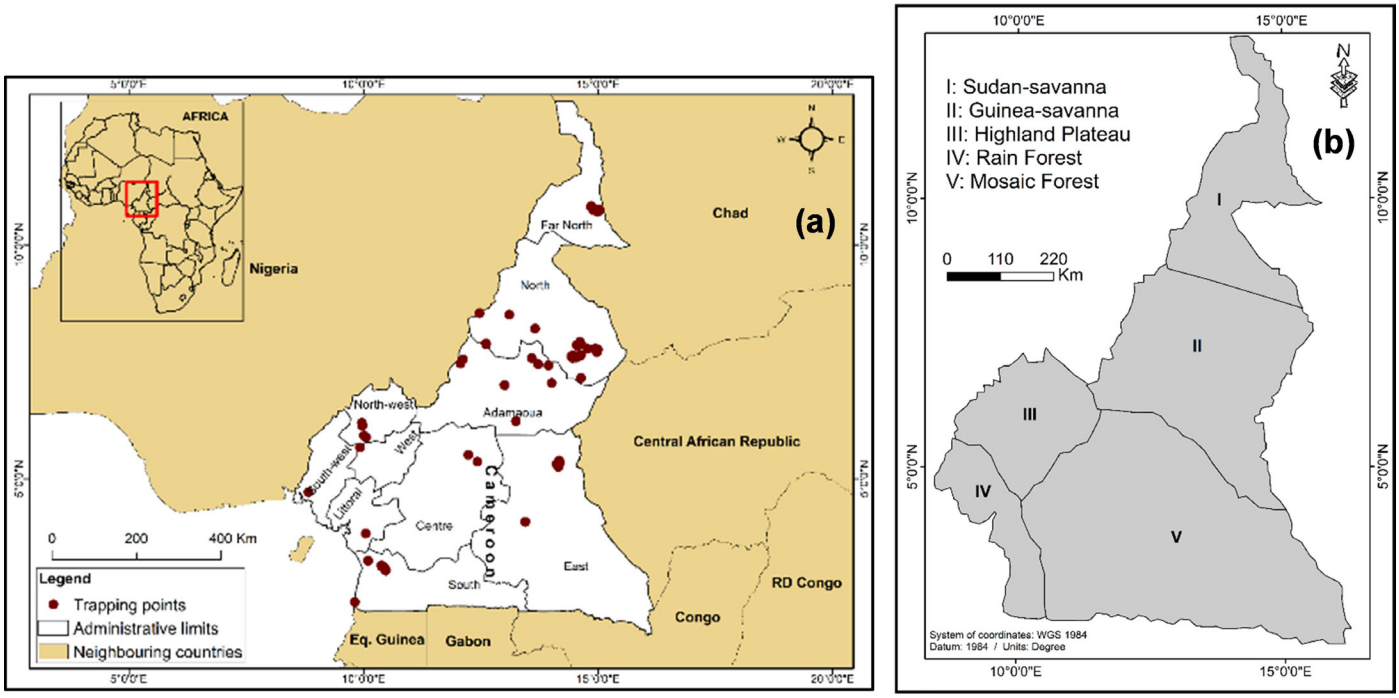

Figure 1 Location of study area. (a) Map of Cameroon showing study regions and sampling sites; (b) Agro-ecological Zones.

Table 1 Summary table for the description of trapping sites, trap types, trap numbers, time interval and trapping period in the main AEZs of Cameroon.

SN	AEZ	Administrative region	Major cattle rearing regions	Characteristics of trapping points	Trap type	N° of Traps	Time interval	Trapping period
I	Sudan savanna	Far North	x	Marshy land around livestock drinking and grazing spots, Open grass Savanna and Gallery forest	Nzi	20	20th of March to 6th of April 2015	Traps were set at 8 am and emptied at 6 pm.
		North	x	Game Reserve, River banks and livestock grazing spots	Vavoua, Biconical and Nzi	39	2nd of March to 8th of March, 2015	Traps set at 8 am and emptied at 6 pm.
II	Rainforest	Littoral		Open forest, cattle grazing spots, humid and closed forest with tall canopy trees	Vavoua, Biconical and Nzi	18	6th of January to 9th of May 2017	Traps were set at 6 am and emptied at 6 pm.
III	Guinee savanna	Adamawa	x	Lake Djalingo with open grass Savanna representing cattle grazing spots, forest- Savanna mosaic, River vina banks, and gallery forest	Vavoua, Biconical, Nzi and MVT	56	4th of October 2016 to 18th of June 2017	Traps were set at 7 am and emptied at 6 pm.
IV	Mosaic forest	East		Gallery forests, cattle corrals and river banks representing animal drinking spots	Nzi and Vavoua	6	15th of February to 26th and 15th of May to 26th 2017	Traps were emptied after 24 hrs of exposition.
		Center		Livestock settlement areas	Vavoua	30	24th October to 1st November 2017	Traps were set at 10 am and emptied at 4 pm.
		South		Human settlement areas, forest and mangrove	Vavoua	30	9th to 13th of July 2016	Traps were checked every 7 am.
V	Highland plateau	North West	x	Open grass savanna-forest mosaic representing cattle grazing spots	Biconical	5	11th of December 2013 to 25th of May 2014	Traps were set at 7 am and emptied at 6 pm.

SN: site number; AEZ: agro-ecological zone; SODEPA: livestock production and exploitation corporation; x: major cattle rearing regions.

Fly identification

All specimens preserved in ethanol (Votýpka et al., 2019) were identified by the first author by using a stereo microscope (Carl Zeiss™ STEMI 2000-C). The identification of *Stomoxys* was conducted following the morphological identification key from Zumpt (1973). The *Stomoxys* specimens are kept in insect boxes in the entomology unit of the Laboratoire d'Écologie des Maladies Transmissibles (LEMAT) Libreville, Gabon.

Determination of abundance

The abundance of trapped *Stomoxys* was defined as their apparent density, stated as the number of *Stomoxys* per trap per day (s/t/d) (Sevidzem et al., 2015) as follows:

$$ADT = \frac{NSC}{NT \times ND} \quad (1)$$

Where:
ADT: Apparent density,
NSC: Number of *Stomoxys* captured,
NT: Number of traps,
ND: Number of trapping days

Statistical analysis

Stomoxys data was completed in Access (Microsoft) data bases and joined to the trap point shape files produced from gpx files from the GPS handset in ArcMap™ version 10.1, Geographic Information System (GIS) software (Environmental Systems Research Institute, USA). Data was analysed using the R statistical software (RStudio Team, 2020). For the comparison test, a Kruskal-Wallis non-parametric test for the initial detection of a significant difference in the *Stomoxys* abundance among regions was used, followed by the Dunn *post-hoc* test with Bonferroni correction, to account for multiple group comparison. The significant level of all the tests was set to $p<0.05$.

Results

Stomoxys species occurrence and distribution across administrative regions and AEZs

The present survey led to the collection of 77804 *Stomoxys* specimens from 606 trapping points in 28 sites in eight regions of the five AEZs of Cameroon. *Stomoxys* spp. were identified as *Stomoxys niger niger*, *S. calcitrans*, *S. niger bilineatus*, *S. xanthomelas*, *S. omega*, *S. transvittatus*, *S. inornatus* and *S. sitiens*. *Stomoxys niger niger* and *S. calcitrans* were the only taxa present in most of the regions (five to seven out of eight), while *S. transvittatus* and *S. inornatus* were only encountered in the Guinean savanna of the Adamawa Plateau. *Stomoxys sitiens* was very rare, only identified in the Sudan savanna of the North region (Table 2).

At the species level, *S. calcitrans* is found in the Sudan savanna of the North region in the livestock-human hunting grounds-wildlife interface (Figure S1A). This species occurs in the Guinean savanna of Adamawa in cattle settlement areas, in the rain forest zones of the littoral, in the Highland Plateau of the North West around cattle grazing land, and the mosaic forest of the South around the Game Reserve (Campo Ma'an National Park). *Stomoxys omega* was encountered in the Guinean savanna of the Adamawa Plateau; in the Rainforest near the Game Reserve, and the Mosaic forest of the Littoral and South regions respectively (Figure S1B). *Stomoxys sitiens* was only found in the human hunting grounds of the Sudan savanna of the North region (Figure S2A). *Stomoxys xanthomelas* was found in the gallery forests of the Guinean savanna in Adamawa; in the palm oil plantation areas of the Littoral region, and in the mangrove sites of the South region (Figure S2B). *Stomoxys niger niger* was encountered in all the AEZs (Figure S3A). *Stomoxys niger bilineatus* was found in the Sudan savanna of the North around cattle grazing areas, in the Guinean savanna of the Adamawa region around stables, in the Mosaic savanna of the East and South regions (Figure S3B). *Stomoxys transvittatus* (Figure S4A) and *Stomoxys inornatus* (Figure S4B) were only caught in the cattle settlement areas of the Adamawa region (Table 2).

Abundance

The overall mean apparent density of *Stomoxys* captured from the different regions of Cameroon was 3.5 s/t/d. The highest ADT range of 101 to 200 s/t/d was recorded in the Sudan savanna of the Far North region. In the other regions the ADT range was 0 to 50 s/t/d (Fig. 2).

There was a statistically significant difference (Kruskal-Wallis Chi-squared = 103.51, df = 4, p<0.001) in the ADT of *Stomoxys* in the regions. Additionally, the Dunn post-hoc test showed significant differences between pairwise AEZ comparisons (Table 3; Table S1).

Discussion

The *Stomoxys* fauna of Cameroon comprises eight taxa, with *Stomoxys niger niger* and *Stomoxys calcitrans* having the highest abundance and distribution across the eight administrative regions and in key AEZs of Cameroon. It is known that of the 18 known species of *Stomoxys* flies, *S. calcitrans* is cosmopolitan, and studies on its biogeography indicates that it followed human beings during their peregrinations around the world; from tropical to continental climates (Zumpt, 1973; Dsouli-Aymes et al., 2011). Its origin was probably the Oriental region (Zumpt, 1973; Dsouli-Aymes et al., 2011) and not the Ethiopian region.

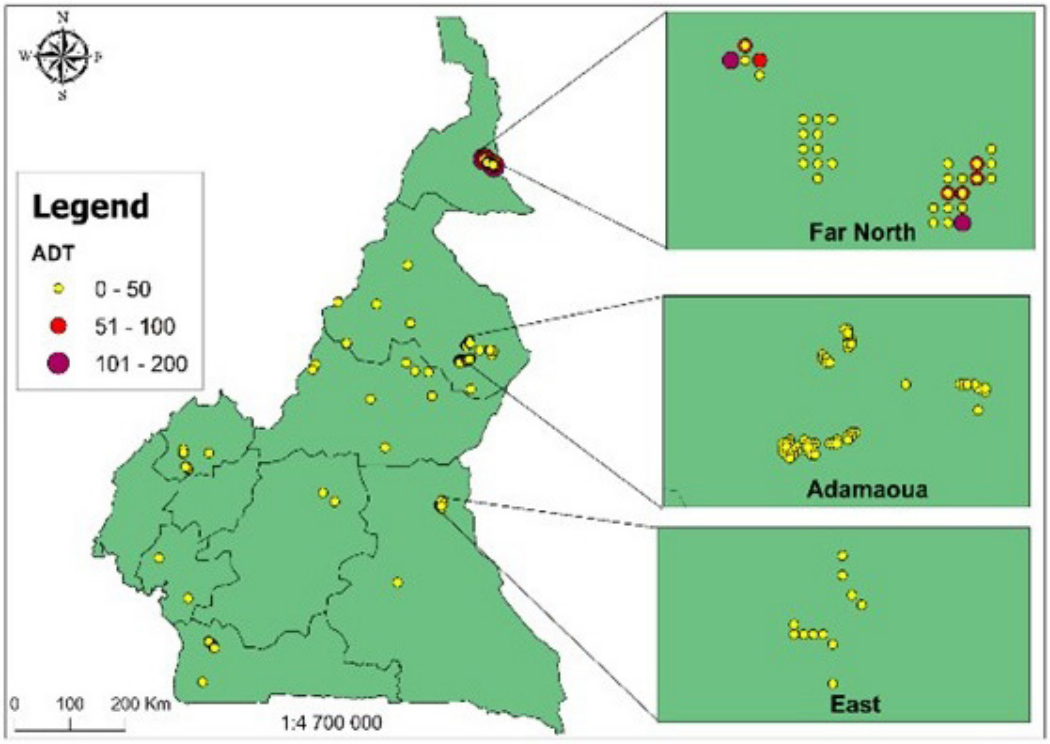

Figure 2 *Stomoxys* apparent density in the eight regions of Cameroon.

Table 2 Species number and composition in the main AEZs and regions.

AEZ	Region	n	Sc	Si	Snb	Snn	So	Ss	St	Sx
Guinean Savanna	Adamawa	7	x	x	x	x	x		x	x
Sudan Savanna	North	4	x		x	x		x		
	Far North	1				x				
Mosaic Forest	East	4			x	x	x			x
	South	5	x		x	x	x			x
	Center	1								
Rain Forest	Littoral	4	x			x	x			x
Highland Plateau	North West	2	x			x				

AEZ: agro-ecological zone; Snn: Stomoxys niger niger; Sc: Stomoxys calcitrans; Snb: Stomoxys niger bilineatus; Sx: Stomoxys xanthomelas; So: Stomoxys xanthomelas; St: Stomoxys transvittatus; Si: Stomoxys inornatus; Ss: Stomoxys sitiens. (x): Stomoxys spp. Presence; n: number of species.

Table 3 Comparison of abundance of *Stomoxys* between agro-ecological zones/administrative regions.

Agro-ecological zone	Region	min	max	median	mean	Significance
Guinea savanna	Adamawa	1	30	2	5.6	ab
Highland Plateau	North West	0	5	1	1.7	ac
Mosaic forest	East, South, Center	0	13	1	2.5	ac
Sudan savanna	Far North	0	200	6.5	22.2	b
	North	0	3	0	1.0	c

The Rainforest ecozone was dropped from the comparison due to a small sample size (2 observations). Matching letters means that there is no significant difference at the 0.05 threshold.

Also, the frequency of *S. niger niger* in the present collection was expected, considering Zumpt (1973) report of the widespread distribution of this species in tropical African settings. The frequent occurrence of both *S. calcitrans* and *S. n. niger* was also reported in Cameroon (Sevidzem et al., 2016; Mounioko et al., 2017; Fongho et al., 2019; Hiol et al., 2019; Sevidzem et al., 2022), in neighbouring Gabon (Bitome-Essono et al., 2015; Mavoungou et al., 2017; Mounioko et al., 2018; Sevidzem et al., 2019a), and neighbouring Nigeria (Ahmed et al., 2005).

The eight taxa (*S. calcitrans*, *S. niger niger*, *S. niger bilineatus*, *S. xanthomelas*, *S. inornatus*, *S. transvittatus*, *S. sitiens* and *S. omega*) recorded for Cameroon was higher than that reported in studies from neighbouring Gabon (*S. calcitrans*, *S. niger niger*, *S. niger bilineatus*, *S. xanthomelas*, *S. inornatus*, *S. transvittatus*, and *S. omega*) and Nigeria (*S. calcitrans* and *S. niger niger*), for which seven and two taxa, respectively, were reported. *Stomoxys sitiens* was a rare finding in the present study and has not yet been reported anywhere in the sub-region. However, apart from its earlier identification in the North region of Cameroon by the first author in 2015 (Sevidzem et al., 2016), other reports indicate that *S. sitiens* has been recorded from many areas in the Ethiopian region from Gambia to South Africa and Egypt, however it is still rare in these collections (Dsouli-Aymes et al., 2011). Additionally, it occurs in the Oriental region from India to the Philippines, but again, specimens are rare, as found in studies from Africa (Zumpt, 1973). The occurrence of this species in the Sudan savanna of the North region of Cameroon is likely due to the long period of study (three years of exhaustive and systematic trapping), combination of different trap types, the presence of cattle and wild animal hosts, and the conducive environmental conditions of this region, which, presumably, favors its development and survival. The Sudano-Sahelian climate has a mean annual monthly temperature of 30°C and humidity of 84% that is closer to the ideal hatching mean temperature (32°C) and mean humidity (90%) as well as the ideal adult emergence mean temperature (27°C) and mean humidity (70%) for *S. sitiens* reported by Issimov et al. (2020).

Regarding the species composition and abundance within the regions of the different AEZs, the Guinean savanna of the Adamawa region recorded the highest species number (seven species), which aligns with a previous study on bionomics of symbovine dipterids that reported high diversity of *Stomoxys* on cattle in the Adamawa region (Lendzele et al., 2019). However, the highest abundance range was recorded in the Sudan savanna of the Far North region. The highest species diversity and density in the Adamawa and Far North regions, respectively, could be explained by the fact that these regions are primary livestock rearing areas of Cameroon (MINEPIA, 2013), thereby providing the host (cattle) and fertile habitat (decaying grass mixed with urine and faeces) for the development and proliferation of *Stomoxys*. Additionally, the Sudano-Guinean climate of Adamawa has an average annual monthly temperature of 29°C that is conducive for the flight activity of adult *Stomoxys*. The study of Ahmed et al. (2005) in neighboring Nigeria established that the ambient temperature to get high catch of biting flies including *S. calcitrans* and *S. n. niger* was 22.8-24.1°C. The study of Sevidzem et al. (2019c) reported that the diurnal temperature and air humidity range conducive for the flight activity of *Stomoxys* in the Sudano-Guinean climate of Adamawa is 31°C and 50%, respectively. The abiotic and biotic conditions of the Adamawa region clearly indicates that *Stomoxys* can survive and develop in this milieu, reason for high species richness in this region.

This study had some limitations as trapping was not conducted in both seasons for all the study regions, and there were limited traps (so different traps were used). Future studies will include seasonal entomological and epidemiological surveys and trapping will be conducted using same trap-type and numbers for the different regions in order to get comparable data for Cameroon.

Conclusions

Eight taxa (*Stomoxys niger niger*, *S. calcitrans*, *S. niger bilineatus*, *S. omega*, *S. xanthomelas*, *S. inornatus*, *S. transvittatus* and *S. sitiens*) comprise the *Stomoxys* fauna of Cameroon with a heterogenous distribution in the eight studied regions. *Stomoxys calcitrans* and *S. niger niger* were found in most of them, while *Stomoxys sitiens* was identified only in the Sudan savanna of the North region. The Guinean savanna of the Adamawa plateau recorded the highest number (seven out of eight) but the Sudan savanna of the Far North region had the highest abundance range (101 to 200 s/t/d). This study provided relevant information of species abundance and distribution to further understand the potential health risk for Cameroon livestock populations. Future investigations, regarding the *Stomoxys* group, should be completed to identify and characterize the pathogens they harbor and to establish their potential epizootiological role in the different regions of Cameroon.

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Data statement

The data presented in this study are available on request from the corresponding author.

Conflicts of interest

The authors declare no conflict of interest.

Author contribution statement

SSL, ABK, MA, MMMM, RMN, JFM conceived and supervised the study. SSL and ABK undertook collection of field data and SSL identified all the species. AAK, ABK and SSL constructed all the *Stomoxys* distribution maps. SRM conducted statistical analysis. ABK, SSL and SRM interpreted the data and wrote initial manuscript while HB revised the subsequent drafts. All authors read and approved the final manuscript for publication and remain in agreement on all aspects of the work.

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Supplementary material

The following online material is available for this article:
Figure S1 - Distribution maps of S. calcitrans (A) and S. omega (B).
Figure S2 - Distribution maps of S. sitiens (A) and S. xanthomelas (B).
Figure S3 - Distribution maps of S. niger niger (A) and S. niger bilineatus (B).
Figure S4 - Distribution maps of S. transvittatus (A) and S. inornatus (B).
Table S1 - Supporting information showing the p-values for each comparison across AEZs.