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STUDIES ON THE SPECIES COMPOSITION AND RELATIVE ABUNDANCE OF MOSQUITOES OF MPIGI DISTRICT, CENTRAL UGANDA

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Abstract

Prediction of arboviral disease outbreaks and planning for appropriate control interventions require knowledge of the mosquito vectors involved. Although mosquito surveys have been conducted in different regions of Uganda since the mid 30's such studies have not been carried out in Mpigi District. In October 2011, we conducted mosquito collections in Mpigi district to determine species composition and relative abundance of the different species. The survey was conducted in four villages, Njeru, Ddela, Kiwumu and Nsumbain Kammengo sub-county, Mpigi district, Uganda. CDC light traps baited with dry ice (carbon dioxide) were used to capture adult mosquitoes. A total of 54,878 mosquitoes comprising 46 species from eight genera were collected. The dominant species at all sites was *Coquilletidia (Coquilletidia) fuscopennata* Theobald (n=38,059, 69%), followed by *Coquillettidia (Coquillettidia) metallica* Theobald (n=4,265, 7.8%). The number of species collected varied from 17 in the genus *Culex* to 1 in the genus *Lutzia*. Of the 46 species identified, arboviruses had previously been isolated from 28 (60.9%) suggesting a high potential for arboviral transmission and/or maintenance in Mpigi District.

Keywords

mosquito; Mpigi; species composition; Uganda

Introduction

Mosquitoes of Uganda were extensively investigated between the mid-1930s and the early 1970s ^[1]. During that period more than 220 mosquito species were recorded and approximately 20% were implicated in arbovirus transmission ^[2]. Because of these investigations, Uganda is currently a known hotspot for emergence of arboviruses of public health and veterinary importance ^[3, 4]. Although earlier studies have described species composition, distribution, behavior and preferred habitat of mosquitoes in Uganda, most of these studies were conducted >40 years ago and only in limited areas of the country ^[1, 5–17]. Such information has been more recently updated by Mutebi *et al* ^[1] for western Uganda and Kaddumukasa *et al*. ^[18] for Zika Forest. In the intervening years arthropod surveillance

activities in Uganda have mainly been limited to collections related to specific taxonomic questions or to monitoring vectors during outbreaks, such as the O'nyong-nyong virus(ONNV) outbreak in 1996 ^[19, 20]. Therefore, more generalized mosquito surveys are needed to develop distribution maps of medically significant vector species to facilitate prediction and planning for appropriate control strategies for arboviral outbreaks.

We report here a description of mosquitoes of Mpigi district in central Uganda, an area for which mosquito species composition and abundance data is not available. This study was part of the arbovirus surveillance program recently established at the Uganda Virus Research Institute (UVRI) in Entebbe, Uganda, in collaboration with the US Centers for Disease Control and Prevention. This study is a first step toward surveillance for arboviruses of public health and veterinary importance in Mpigi District.

Materials and Methods

Collection sites

Mosquito collections were conducted in Kammengo Sub County, Mpigi District in central Uganda (Fig. 1). This district is approximately 37 km west of Kampala and borders Wakiso District to the North and East, Kalungu District to the South, Butambala District to the West and Mityana District to the North-West. Mosquitoes were collected from four villages: Njeru (Lat 0.113533, Lon 32.22035), Ddela (Lat 0.106433, Lon 32.2248), Kiwumu (Lat 0.086867, Lon 32.234183) and Nsumba (Lat 0.083467, Lon 32.206917) (Fig. 1). The distance between the collection sites ranged from 0.9 km (Njeru to Ddela) to 3.6 km (Njeru to Nsumba) with a median distance of 3.1 km. The altitude at the study site was between 1166m and 1194m above sea level. The collection sites were in plantations and small gardens close to residential areas and near wetlands that stretch to the shores of Lake Victoria. The primary occupation of the local population in the area is subsistence farming; mostly small scale banana and coffee plantations, vegetable fields and/or gardens and limited rearing of cattle, goats, ducks, chickens and other farm animals.

Mosquito collection

Mosquito collections were made from October 15 through 24, 2011. CDC light traps baited with dry ice (carbon dioxide) were suspended on tree branches in coffee plantations, banana plantations and bushes near homes. Twenty five CDC light traps were hung at each site at approximately 5:00pm, left overnight for about 14 hours and collected each morning at approximately 8:00am. Collections were made at each site for 2 nights. The contents of the traps were sorted into cryovials and transported frozen in liquid nitrogen dry shippers to the UVRI laboratory in Entebbe where they were stored at -80° C until identification.

Mosquitoes were identified to species using the morphological keys of Edwards ^[21], Huang ^[22], Jupp ^[23], Gilles and De Mellion ^[24], and Gilles and Coetzee^[25]. Specimens were pooled by species, sex, feeding status and parity and stored at -80° C for virus isolation studies. Voucher specimens were retained for each species for future reference and identification consultation.

Diversity Indices

Species richness and diversity were calculated for each collection location. While species richness is the number of mosquito species at each location, species diversity takes into account both species richness and the relative abundance of each species. Species diversity was estimated by using the Simpson Index^[26]. The Simpson Index was calculated as $D=\Sigma n(n-1)/N(N-1)$ where n= the total number of mosquitoes of a particular species and N=the total number of mosquitoes of all species in each collection. We also report the Simpson's Index of Diversity (1-D), which is interpreted as the greater the index value, the greater the sample diversity.

Results and Discussion

A total of 54,878 mosquitoes belonging to 9 genera and 45 species were identified. The number of mosquitoes collected from the individual sites was: Njeru (n=16,403), Ddela (n=16,818), from Kiwumu (n=12,099) and Nsumba (n=9,555). Table 1 lists the species captured by collection site. Twenty (44.4%) of the 45 species were captured at all four collection sites which suggests differences in species composition between the sites despite their close proximity (Table 1). Recently Mutebi et al. ^[1] reported differences in species composition between the adjacent sites of Mweya and Maramagambo in western Uganda which suggested that the geographic distance between sites may not be a major determining factor. However, it is important to note that Maramagambo is a forest ecosystem whereas Mweya is an open grassland and therefore the differences in species composition between these two sites may be attributed to differences between the ecosystems. In the present study, 98.5% of the collection belonged to species found at all four sites; the majority of the species that were not captured at all four sites were captured in low numbers. This was most likely due to low abundance or because the species were not strongly attracted to CO₂-baited light traps. Species diversity for the four collection sites, estimated by using the Simpson Index/Index of Diversity which takes into consideration the relative abundance of mosquitoes in each species, ranged from 0.42 - 0.53 (Table 1)^[26]. These values suggest that overall the four collection sites supported similar species diversity.

Figure 2 summarizes the species richness or number of species in each genus collected at the four sites. The highest number of species was in the genus *Culex* at all four sites (Figure 2). At three sites, Ddela, Kiwumu and Nsumba, the second highest number of species was in the genus *Coquillettidia* and the third in the genus *Aedes* (Figure 2). At Njeru, the second highest number of species was in the genus *Aedes*, and the third in genus *Coquillettidia* (Table 2). Overall species richness by genus ranged from 37.0% in the genus *Culex* (37% of all species collected were in the genus *Culex*) to 2.2% in *Hodgesia* and *Mimomyia* (Fig. 2). The genus *Culex* was highest in species richness comprising 17 species (n=5,548, 10.1% of the total collection), followed by the genus *Aedes* (n=611, 1.1%) with 9 species (19.6%) (Table 1, Fig 2). Although the genus *Coquillettidia* had the highest number of mosquitoes captured (n=47,697, 86.9%), it ranked third in terms of species richness with 8 species richness with 6.5% each of the total species identified (Fig 2). The rest of the species were in

the genera *Anopheles*, *Hodgesia* and *Mimomyia* and had minimal contribution to the total species richness in the area (Fig 2).

The most abundant mosquitoes were in the genus Coquillettidia followed by the genus Culex (Table 1). The predominant species collected was Coquillettidia (Coquillettidia) fuscopennata (Theobald) (n=38,059, 69.4%), followed by Coquillettidia (Coquillettidia) metallica (Theobald) (n=4,265, 7.8%) and Coquillettidia (Coquillettidia) fraseri (Theobald) (n=3,129, 5.7%) (Table 1). Coquillettidia species are often the principal species collected near water/wetlands which are their preferred larval habitats. The ecology of the study area in Mpigi District included freshwater swampland in addition to small scale agricultural landscapes adjacent to human habitation, therefore, the large numbers of Coquillettidia mosquitoes collected was not unexpected. The next most abundant species collected were Culex (Oculeomyia) annulioris Theobald (n=2,883, 5.3%) and Culex (Culex) decens group Theobald and Culex (Culex) invidiosus Theobald)(n=1,187, 2.2%) (Table 1)^[21]. Additional species contributing >0.5% to the collection included *Coquillettidia* (*Coquillettidia*) maculipennis (Theobald) (n=611, 1.1%), Coquillettidia (Coquillettidia) aurites (Theobald) (n=603, 1.1%), Coquillettidia (Coquillettidia) cristata (Theobald) (n=480, 0.9%), Culex (Culiciomyia) cinereus (Theobald)(n=462, 0.8%), Aedes (Neomelaniconion) circumluteolus (Theobald) (n=401, 0.7%), Mansonia (Mansonioides) africana nigerrima Theobald (n=399, 0.7%) and Coquillettidia (Coquillettidia) pseudoconopas (Theobald) (n=296, 0.5%) (Table 1).

Of the 46 mosquito species collected in this study arboviruses of medical and veterinary importance had previously been isolated from 28 (60.9%) suggesting roles in the transmission or the maintenance of these arboviruses. This shows a high potential for maintenance and transmission of arboviruses of medical and veterinary importance in Mpigi District. A comprehensive list of arboviruses that have been isolated from mosquito species collected in Uganda has been published by Mutebi *et al.* ^[1]. For many of these species it is unknown whether they play a significant role in epidemic or enzootic virus transmission. However, certain species in the collection are known to be epidemic/enzootic vector species including Aedes (Stegomvia) aegypti (L.), a principal vector of Yellow fever virus (YFV) ^[27, 28], Chikungunya virus ^[29], Dengue viruses^[30] and Zika virus ^[31]. Similarly, members of the Aedes (Stegomyia) simpsoni group have been shown to be epidemic vectors of YFV ^[32, 33] and members of the Anopheles (Cellia) gambiae group are vectors of ONNV^[20, 34]. Additionally epidemic and epizootic transmission of Rift Valley fever virus has been implicated for several species collected in Mpigi district including Aedes (Neomelaniconion) mcintoshi/circumluteolus, Mansonia (Mansonioides) africana (Theobald), Mansonia (Mansonioides) uniformis (Theobald), Culex (Culex) pipiens (L.), Culex (Culex) quinquefasciatus Say, Culex (Oculeomyia) annulioris Theobald and Culex (Oculeomyia) poicilipes Theobald^[35–40]. Some known disease vectors identified in this collection were not captured at all four sites suggesting non-uniform distribution of these vectors in the study sites and an uneven risk of disease transmission across the region. For example members of the Anopheles (Cellia) gambiae group were only detected at Ddela and not at any other study site (Table 1). Similarly, Culex (Culex) pipiens (L) was only detected at Ddela (Table 1). Interestingly Culex (Culex) quinquefasciatus Say, which is closely

related to *Cx. pipiens*, was only detected at Nsumba and not at any other study site (Table 1). Both *Cx. pipiens* and *Cx. quinquefasciatus* which are commonly captured in C02-baited light traps were found in low numbers suggesting that the densities of these two species were low at the study sites. *Cx. pipiens* and *Cx. quinquefasciatus* are commonly associated with human residences (domestic species). Since collections in this study were conducted in domestic and peridomestic areas, our observations suggest that the risk of *Cx. pipiens* and *Cx. quinquefasciatus*-transmitted pathogens is low in this area.

To our knowledge this is the first documented account of the mosquito fauna of Mpigi District in central Uganda. All of the 45 species identified had previously been identified elsewhere in Uganda and do not represent new introductions to the country. Species richness in this area was high and >60% of the mosquito species collected have been previously documented as potential arbovirus vectors. Increased human activity and encroachment in to undeveloped areas in this region presents a potential risk for exposing humans to arboviral diseases. Historical records show that Uganda has experienced numerous outbreaks of arboviral disease including YFV and ONNV^[20, 41] and many known arboviruses were first isolated from Uganda^[3, 4]. Further study is necessary to determine the seasonal variation in mosquito species composition and abundance in this region. Additionally, virus isolation from collected mosquitoes will be conducted to identify arboviruses that are currently circulating in the area. Such information will be invaluable in predicting and controlling potential outbreaks of arboviral disease in this region.

Acknowledgments

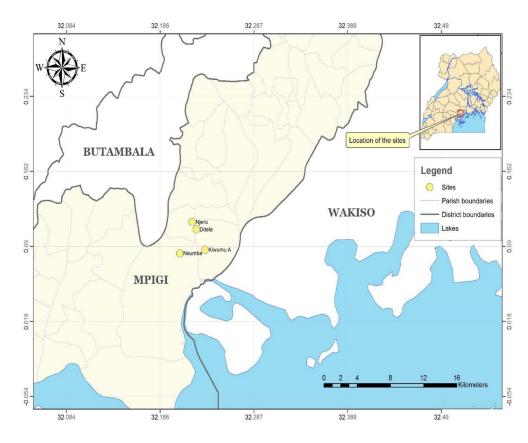
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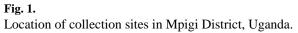
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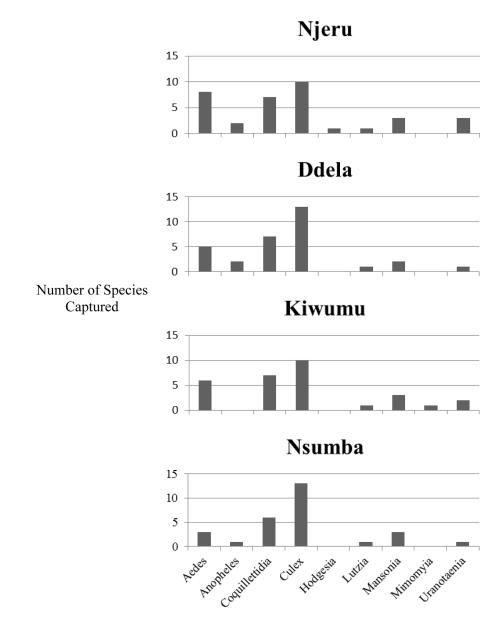
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Mosquito species collected in Mpigi District, central Uganda, in October 2011.

	Cubcound	Current				ź	unber co	Number collected (%)				
Genus	Subgenus	Species	Nj	Njeru	Dc	Ddela	Kiw	Kiwumu	Nsu	Nsumba	ЧI	All sites
Aedes	Aedimorphus	argenteopunctatus (Theobald)	3	(0.02)							3	(0.01)
		cumminsii (Theobald)	26	(0.16)							26	(0.05)
		phyllolabis Edwards	25	(0.16)	10	(0.06)	10	(0.08)	10	(0.10)	55	(0.10)
		tarsalis (Newstead)					3	(0.02)			3	(0.01)
	Finlaya	ingrami Edwards	1	(0.01)	1	(0.01)					7	(0.00)
	Neomelaniconion	circumluteolus (Theobald)	136	(0.85)	112	(0.67)	113	(0.93)	40	(0.42)	401	(0.73)
		<i>mcintoshi</i> Huang	19	(0.12)	25	(0.15)	25	(0.21)	22	(0.23)	91	(0.17)
	Stegomyia	aegypti (L.)	1	(0.01)	9	(0.04)	8	(0.07)			15	(0.03)
		<i>simpsoni</i> group ^a	1	(0.01)			3	(0.02)			б	(0.01)
		species			4	(0.02)	8	(0.07)			12	(0.02)
Anopheles	Anopheles	coustani Laveran	S	(0.03)	7	(0.04)			1	(0.01)	13	(0.02)
		implexus (Theobald)	10	(0.06)							10	(0.02)
	Cellia	$gambiae\ { m group} b$			26	(0.15)					26	(0.05)
Coquillettidia	Coquillettidia Coquillettidia	aurites (Theobald)	186	(1.16)	187	(1.11)	155	(1.28)	75	(0.78)	603	(1.10)
		fraseri (Theobald)	774	(4.82)	1292	(7.68)	476	(3.93)	587	(6.14)	3129	(5.70)
		fuscopennata (Theobald)	11508	(71.73)	11412	(67.86)	9175	(75.83)	6444	(67.44)	38539	(70.23)
		maculipennis (Theobald)	238	(1.48)	135	(0.80)	148	(1.22)	90	(0.94)	611	(1111)
		metallica (Theobald)	1142	(7.12)	1754	(10.43)	689	(5.69)	674	(7.05)	4259	(7.76)
		pseudoconopas (Theobald)	52	(0.32)	134	(0.80)	57	(0.47)	53	(0.55)	296	(0.54)
		versicolor (Barraud)	49	(0.31)	124	(0.74)	56	(0.46)			229	(0.42)
		species	15	(60.0)			16	(0.13)			31	(0.06)
Culex	Culex	antennatus (Becker)	52	(0.32)	29	(0.17)	19	(0.16)	41	(0.43)	141	(0.26)
		decens group ^c	176	(1.10)	373	(2.22)	261	(2.16)	377	(3.95)	1187	(2.16)
		neavei Edwards			6	(0.05)	L	(0.06)	80	(0.84)	96	(0.17)
		ornatothoracis Theobald	56	(0.35)	44	(0.26)	42	(0.35)	45	(0.47)	187	(0.34)
		perfuscus Edwards							11	(0.12)	11	(0.02)

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	Curberry C	Carolina Carolina				N	umber co	Number collected (%)	()			
sulla	Sungenus	openes	Ŋ	Njeru	Dd	Ddela	Kiw	Kiwumu	Nsi	Nsumba	ЧI	All sites
		pipiens (L.)			2	(0.01)					2	(0.00)
		quinquefasciatus Say							7	(0.02)	2	(0.00)
		trifilatus Edwards			1	(0.01)			19	(0.20)	20	(0.04)
	Culiciomyia	cinerellus Edwards			1	(0.01)					1	(0.00)
	Culiciomyia	cinereus Theobald	257	(1.60)	117	(0.70)	21	(0.17)	67	(0.70)	462	(0.84)
		nebulosus Theobald	23	(0.14)	13	(0.08)			116	(1.21)	152	(0.28)
	Eumelanomyia	insignis (Carter)	22	(0.14)	37	(0.22)	24	(0.20)	30	(0.31)	113	(0.21)
		rubinotus Theobald	50	(0.31)	29	(0.17)	103	(0.85)	66	(0.69)	248	(0.45)
	Kitzmelleria	<i>moucheti</i> Evans			5	(0.03)	-	(0.01)	4	(0.04)	10	(0.02)
	Oculeomyia	annulioris Theobald	1212	(7.55)	746	(4.44)	439	(3.63)	486	(5.09)	2883	(5.25)
		amulioris consimilis Newstead	13	(0.08)			20	(0.17)			33	(0.06)
		poicilipes Theobald	33	(0.02)							33	(0.01)
		species							14	(0.15)	14	(0.03)
Hodgesia	Hodgesia	sanguinae Theobald	17	(0.11)							17	(0.03)
		species	32	(0.20)							32	(0.06)
Lutzia	Metalutzia	tigripes de Grandpre & de Charmoy	26	(0.16)	36	(0.21)	18	(0.15)	28	(0.29)	108	(0.20)
Mansonia	Mansonioides	africana (Theobald)	4	(0.02)			10	(0.08)	2	(0.02)	16	(0.03)
		africana nigerrima Theobald	178	(1.11)	90	(0.54)	68	(0.56)	63	(0.66)	399	(0.73)
		uniformis (Theobald)	65	(0.41)	32	(0.19)	32	(0.26)	36	(0.38)	165	(0.30)
Mimomyia	Mimomyia	hispida (Theobald)					7	(0.02)			5	(0.00)
Uranotaenia	Pseudoficalbia	mashonaensis Theobald	7	(0.01)			7	(0.02)			4	(0.01)
	Uranotaenia	alboabdominalis Theobald	ю	(0.02)							33	(0.01)
		palidocephala Theobald	21	(0.13)	25	(0.15)	63	(0.52)	47	(0.49)	156	(0.28)
		species					26	(0.21)	25	(0.26)	51	(0.0)
Total			16403		16818		12099		9555		54875	
Grand total		54,875										
D			0.51		0.48		0.58		0.47		0.51	
1-D			0.49		0.52		0.42		0.53		0.49	

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^bThe An. gambiae group consist of at least six species An. gambiae s.s. Giles, An. arabiensis Patton, An. quadriannulatus Theobald, An. melas Theobald, An. merus Dönitz, and An. bwambae White (Gilles and Coetzee 1987).

^cThe Culex decens group includes Cx. decens Theobald and Cx. invidiosus Theobald (Edwards 1941).