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Diversity, distribution and abundance of potential rift valley fever vectors in Baringo County, Kenya

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Abstract

Rift Valley fever (RVF) is a zoonotic disease that occurs sporadically in form of outbreaks and is transmitted by diverse mosquito species in different geographic regions. Knowledge on diversity, distribution and abundance of RVF vectors is useful for risk assessment of RVF outbreaks. Diversity, distribution and abundance of RVF vectors from four ecological zones in Baringo County were studied. Four potential RVF vectors, namely *Mansonia uniformis*; *Mansonia africana*; *Culex pipiens* and *Culex univittatus* were among the 26 species identified. Rift Valley Fever vectors were most abundant in lowlands (85.9%); riverine (9.1%); midland and highland combined (5%). Diversity indices were higher in the riverine ($H' = 1.65$) and midland ($H' = 1.64$) than lowland ($H' = 1.429$) and highland ($H' = 1.229$). Area-specific vector distribution and abundance data generated from this study can be incorporated into the national RVF contingency Plan as part of an improved preparedness and early response to RVF outbreaks.

Keywords: RVF, Vectors, Diversity, Distribution, Abundance, Baringo

Introduction

Mosquitoes are the most important group of biting dipteran ^[1] which transmit variety of diseases including Rift Valley fever (RVF). Rift Valley fever is an arboviral zoonotic disease that is endemic in the African continent but has also spread to the Arabian Peninsula where it was first reported in the year 2000 ^[2]. It was first reported in 1912 in the Rift Valley province of Kenya and remained confined there until 1960's when it spread to other regions of Kenya ^[3]. Eleven national RVF epizootics have been reported in Kenya since 1950 up to 2007 at intervals ranging between 1-7 years ^[3]. Rift Valley fever leads to significant losses in terms of human illnesses, livestock abortions and death ^[4]. Animals are mainly infected through mosquito bites while humans get exposed to the disease through contact with body fluids and tissues of infected animals ^[5]. Investigations during and after the 2006/2007 RVF outbreak in Kenya showed that individuals who handled aborted animal fetuses got severe disease while those who consumed products from sick animals died ^[6].

The geographic expansion of RVF has been increasing with each successive outbreak; with the presence of competent vector species being crucial for virus transmission and establishment in new areas ^[5, 7, 8]. Many species of mosquito vectors are capable of transmitting RVF virus in different countries ^[5, 8, 9] but the dominant species are flood water *Aedes* species such as *Aedes mcintoshi* and *Aedes ochraceus* usually referred to as primary vectors ^[10]. Other mosquito species in the *Culex* and *Mansonia* genera usually referred to as secondary vectors and a few potential species in the genus *Anopheles* also play a great role in the spread of RVF during outbreaks ^[10].

Baringo has been classified as a high RVF risk area in Kenya based on semi-quantitative risk assessment by experts ^[11]. The County is highly heterogeneous in terms of topography and altitude and the local communities are agro-pastoralists. Although several studies have been conducted on mosquito vectors, few have investigated the entire Baringo County. One such study was conducted by Reiter *et al.* who surveyed Kerio valley and highlands during the yellow fever outbreak in 1993 ^[12]. Previous studies have focused on entomological surveys around the lakes in the lowland ^[13-19] therefore vector species diversity and abundance in the entire County to predict RVF risk has not been established after the last outbreak of 2006/2007.

The current study investigates RVF vector diversity, distribution and abundance in diverse areas of Baringo County. Data generated would be indicative of RVF hotspot areas and serve as part of early warning signs for RVF outbreaks in the region.

2. Materials and Methods

2.1 Study Area Description

The study was conducted within Baringo County of Kenya (Figure1), lying between 35.602°E - 36.277°E and 0.541°N - 0.723° N at altitudes ranging between 870 and 2499 m asl. The region is inhabited by Tugen, Ilchamus and Pokot communities who are mainly agro-pastoralists. There are four

lakes within the study area, two of which are permanent (Lake Baringo and Lake Bogoria) and the other two seasonal (Lake 94 and Lake Kamnarok). Most rivers in the area cease to flow during the dry season and are often characterized by pockets of small pools along the riverbed, which provide suitable breeding micro-habitats for mosquito vectors. There also exist dams that form focal points where humans and livestock aggregate to access water especially during the dry season. The mean annual rainfall is about 650mm with temperature ranging between 30 °C to 37 °C. Rift valley fever outbreak occurred once in 2006/2007 and was limited to a few hotspot areas around the L. Baringo and L. 94 [14].

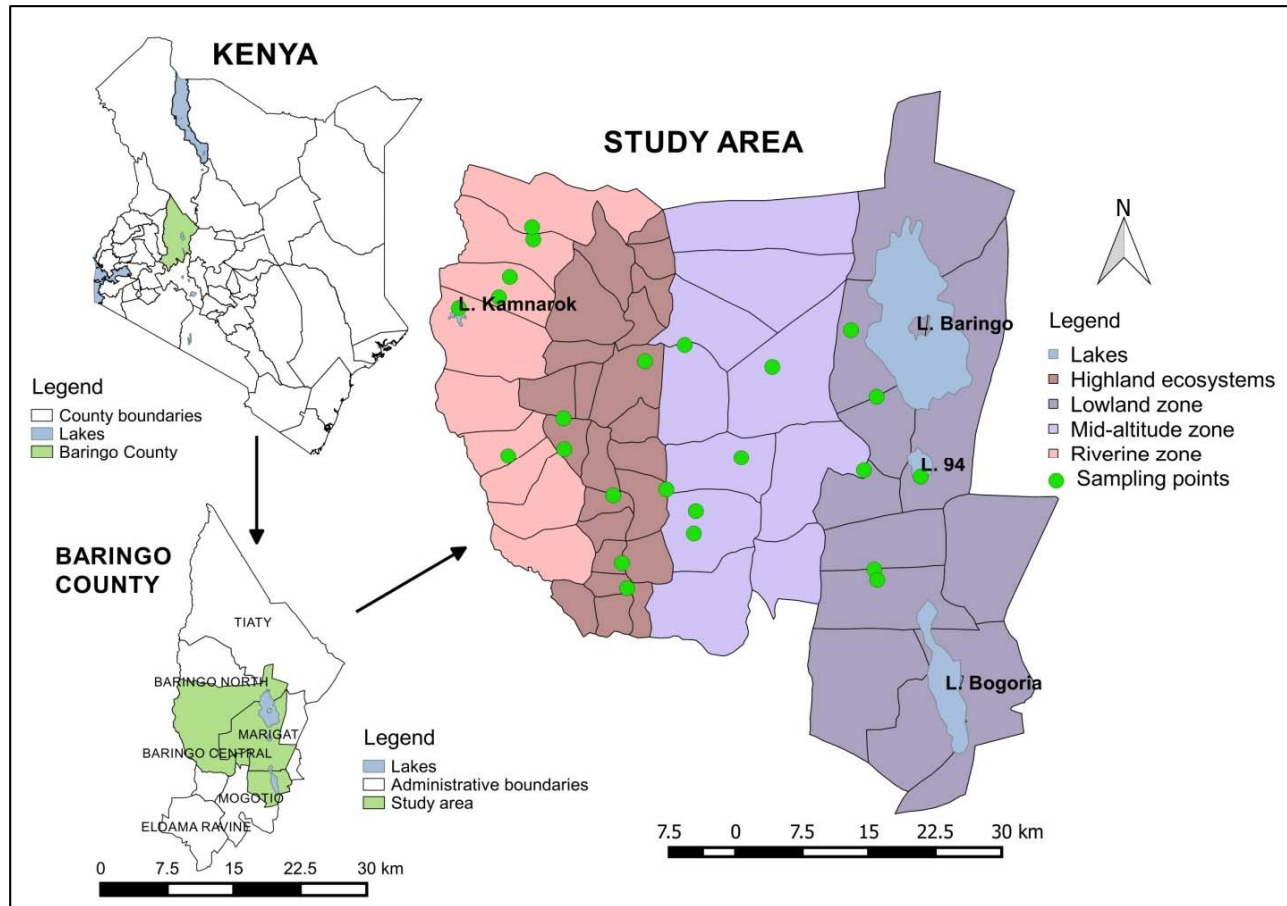


Fig 1: Map of study area showing sampling points in Baringo County

2.2 Sampling design and mosquito identification

The study area was stratified into four zones namely; lowland, riverine, midland and highland based on elevation. Six sampling points were randomly selected from each of the four zones making a total of 24 sites. Indoor and outdoor resting mosquitoes were sampled monthly for a period of 12 months between June 2015 and May 2016. Outdoor mosquitoes were collected using CDC light trap while indoor mosquitoes were sampled by pyrethrum spray collection (PSC). Outdoor sampling was done between 1800 - 0600 hours, a night preceding indoor sampling which was done between 0600 - 0830 hours. All mosquitoes were identified morphologically under the dissecting microscope using taxonomic keys [20, 21].

2.3 Statistical analyses

The relative abundance of the species was expressed as the percentage of the total number of mosquitoes collected. Species diversity was estimated for each zone by Shannon diversity index in PAST software version 2.17c. Negative binomial model was used to determine influence of zone on mosquito abundance. Separate models were fitted for each species of RVF vectors in R version 3.3.1. *Mansonia uniformis* and *Mansonia africana* were combined while *Culex pipiens s.l.* and *Culex univittatus* were analyzed separately.

3. Results and Discussion

3.1 Species diversity and relative abundance

A total of 12,204 mosquitoes were collected and identified to

26 species belonging to 11 genera from both indoor and outdoor places across the four ecological zones (Table 1). Only four genera namely *Aedes*, *Culex*, *Mansonia*, and *Anopheles* have potential and known vectors of RVF. Four species which have been implicated as vectors of RVF were collected and they included; *Mansonia uniformis* (4.4%), *Mansonia africana* (3.3%), which are the principal vectors of RVF in Baringo, *Culex pipiens* s.l. (32.7%) and *Culex univittatus* (2.3%), which are incidental vectors. *Culex pipiens* (76.6%) was the most abundant of the four RVF vectors while *Mansonia uniformis*, *Mansonia africana* and *Cx. univittatus* were less abundant (10.4%, 7.7% and 5.3% respectively). The other *Culex* species which were collected include; *Cx. poicilipes*, *Cx. annulioris*, *Cx. vansomereni*, *Cx. ethiopicus* and *Cx. macfei*. *Culex poicilipes* has been reported as a vector of RVF in Mauritania, Sudan, Senegal and South Africa [5, 8, 9]. Its presence in Baringo County increases the diversity of potential RVF vectors.

Though Baringo County is classified as high risk area for RVF outbreaks [11], only secondary vectors were found. These vectors are known to succeed primary vectors after flooding is over [10]. They then increase in population and incidentally transmit RVF virus to animals. This could imply that they are present during inter-epidemic period but help to spread RVF virus during the outbreak initiated by the *Aedes* primary vectors. Other than some species of *Culex* and *Anopheles*, *Mansonia* species were the most abundant among the other culicines which concurs with previous entomological surveys in Baringo [13, 15].

Species belonging to genus *Aedes* were also collected and these included *Ae. hirsutus*, *Ae. vittatus*, *Ae. metallicus*, *Ae. aegypti* and *Ae. tarsalis*. Throughout the sampling period only 86 individuals of *Aedes* species (0.7%) were collected. However, these species have not been implicated in the transmission of RVF in Baringo. Species of *Aedes* genus were the least abundant probably because they bite outdoors during the day according to previous investigations [22]. Since this study used CDC light traps to collect outdoor mosquitoes at night, the chances of capturing *Aedes* mosquitoes must have been minimized. Alternatively, very few *Aedes* mosquitoes were collected in this study probably due to lack of Carbon dioxide bait normally used with CDC light traps [23]. The *Aedes* primary vectors of RVF in Kenya are *Ae. mcintoshi* and *Ae. ochraceus* commonly referred to as flood water mosquitoes and are believed to maintain the virus transovarially during interepidemic periods. Although these vectors have been reported in small numbers from previous surveys conducted in Baringo [14, 15], they were not collected in this study probably because there was no flooding throughout the sampling period. Another possible explanation for their absence could be because they naturally breed in dambos (shallow depressions which flood during heavy rains) [7] which are not present in Baringo. However, the few

primary vectors of RVF collected during the last outbreak in 2006/2007 in Baringo did not test positive for RVF virus [14]. Since *Aedes* species are suspected to transmit RVF transovarially [7], their low representation in Baringo leaves questions on the source of the virus during the previous outbreak. Nevertheless, other theories propose that RVF virus can be introduced to a region through livestock movement [24]. Baringo being a semi arid County inhabited by pastoralists, the RVF virus during the previous outbreak would have been introduced into the area through animal trade. The *Aedes* species collected in the current study and have been incriminated as vectors of RVF elsewhere included *Ae. vittatus* and *Ae. aegypti* in Saudi Arabia and Sudan respectively [9].

Several other groups of mosquitoes which have not been implicated as vectors were also collected and they include: *Aedeomyia africana*, *Coquillettidia* spp, *Eretmapodite* spp, *Ficalbia* spp, *Hodgesia* spp, *Orthopodomyia* spp and *Uranotaenia* spp. *Anopheles* mosquitoes which included *Anopheles gambiae*, *An. funestus*, *An. coustani* and *An. pharoensis* were also collected. Studies conducted elsewhere have incriminated some anopheline species by isolating RVF virus from wild specimens during outbreaks or by demonstrating infectivity in laboratory experiments [9, 25]. For example, *Anopheles coustani* has been reported as a potential vector of RVF virus in Madagascar after isolating the virus from wild specimens [26]. This species was abundant outdoors in Baringo an indication it could be strongly zoophilic hence potential vector of RVF virus. Species of *Coquillettidia* and *Eretmapodite* have also been mentioned as potential vectors of RVF virus [25, 26]. The presence of these genera in Baringo is an indication of fast spread of RVF in case of an outbreak.

Species diversity indices were higher in the riverine ($H' = 1.65$) and midland ($H' = 1.64$) than lowland ($H' = 1.429$) and highland ($H' = 1.229$). Similar trend was seen in species evenness whereby midland had the highest index ($e^H/S = 0.516$) followed by riverine ($e^H/S = 0.306$). Although lowland had largest number of species count ($S = 20$), the most abundant species accounted for 44.1% thus lowering species evenness to 0.2088 compared to midland with only 10 species but higher evenness of 0.5159 (Table 1). Generally, high numbers of mosquito species were observed in the lowlands and riverine zones.

This is the first time a comprehensive survey of mosquito species including potential vectors of RVF has been conducted in diverse areas of Baringo. The study has revealed species diversity in different ecological zones. Apart from the study conducted by Reiter *et al.* during the 1992/1993 yellow fever outbreak in Kerio Valley (riverine zone in this study), other entomological investigations have been mainly conducted around the lakes region (lowland in this study) [12-15].

Table 1: Mosquito species diversity and abundance in Baringo County

	Total	Lowland	Riverine	Midland	Highland
<i>Aedes aegypti</i>	1	0	0	0	1
<i>Aedes hirsutus</i>	76	76	0	0	0
<i>Aedes metallicus</i>	1	1	0	0	0
<i>Aedes tarsalis</i>	1	0	0	1	0
<i>Aedes vittatus</i>	6	1	5	0	0
<i>Aedomyia africana</i>	146	0	146	0	0
<i>Anopheles coustani</i>	325	296	22	7	0
<i>Anopheles funestus</i>	69	12	57	0	0
<i>Anopheles gambiae</i>	5412	4294	1053	40	25
<i>Anopheles pharoensis</i>	291	277	14	0	0
<i>Coquillettidia spp</i>	39	36	1	2	0
<i>Culex macfieii</i>	14	2	0	0	12
<i>Culex ethiopicus</i>	49	43	0	2	4
<i>Culex pipiens s.l.*</i>	3987	3653	132	4	198
<i>Culex univittatus*</i>	283	167	109	2	5
<i>Culex vansomereni</i>	5	0	0	0	5
<i>Culex annulioris</i>	4	2	2	0	0
<i>Culex poecilipes</i>	3	1	2	0	0
<i>Eretmapodite spp</i>	9	0	1	7	1
<i>Ficalbia flavopicta</i>	36	34	0	0	2
<i>Ficalbia splendens</i>	179	168	11	0	0
<i>Ficalbia uniformis</i>	52	0	51	0	1
<i>Hodgesia spp</i>	2	0	0	0	2
<i>Mansonia africana*</i>	399	297	97	3	2
<i>Mansonia uniformis*</i>	542	358	137	8	39
<i>Orthopodomyia</i>	32	1	31	0	0
<i>Uranotaenia spp</i>	10	10	0	0	0
Taxa S		20	17	10	13
Individuals		9729	1871	76	297
Shannon H		1.429	1.65	1.641	1.229
Evenness e^H/S		0.2088	0.3063	0.5159	0.2629
Berger-Parker		0.4414	0.5628	0.5263	0.6667

Rift valley fever vectors in Baringo County

Mansonia uniformis, *Ma. africana*, *Culex univittatus* and *Cx. pipiens s.l.* which have been incriminated as vectors of RVF were the most abundant species among the culines. Analysis of diversity indices for vector species alone across the ecological zones showed that lowland had the highest Shannon diversity index ($H' = 0.6705$) followed by highland ($H' = 0.5816$). However, species evenness was still higher in the riverine ($e^H/S = 0.9904$) and midland ($e^H/S = 0.9752$)

compared to lowland and highland (Table 2). High shannon diversity index in the lowland would imply that all the vectors are present in relatively large populations, however, low species evenness may point out to outbreaks in hotspot areas with high vector density as it happened in 2006/2007 [27]. On the other hand, the high species evenness in the riverine would imply uniform outbreak throughout the zone due to expansion of RVF vectors to new areas [28].

Table 2: Rift Valley fever vector species diversity indices and abundance

	Total	Lowland	Riverine	Midland	Highland
<i>Culex pipiens s.l.</i>	3987	3653	132	4	198
<i>Culex univittatus</i>	283	167	109	2	5
<i>Mansonia africana</i>	399	297	97	3	2
<i>Mansonia uniformis</i>	542	358	137	8	39
Taxa S		4	4	4	4
Individuals		4475	475	17	244
Shannon H		0.6705	1.377	1.253	0.5816
Evenness e^H/S		0.4888	0.9904	0.8752	0.4472
Berger-Parker		0.8163	0.2884	0.4706	0.8115

3.2 Distribution and abundance of RVF potential vector species across ecologically distinct zones

Mosquito species abundance varied across zones with lowland having the highest abundance (81.3%) followed distantly by riverine (15.6%). Midland had the lowest population of mosquitoes which accounted for only 0.6% of the total

collections (Table 3). The four vector species implicated in RVF virus transmission; *Mansonia africana*, *Ma. uniformis*, *Cx. pipiens* and *Cx. univittatus* were mainly collected from lowlands (85.9%) and riverine (9.1%) with only 5% collected from midland and highland zones combined.

Table 3: Mosquito species collected in the study sites across the four ecological zones in Baringo County, Kenya

Zone	Site	Secondary vectors				Other mosquito species																	Total
		Cx. pip	Cx. uni	Ma. afr	Ma. unif	*Ae. spp	Aed Afr	An. cou	An. fun	An. gam	An. pha	Coq. spp	cx. mac	Cx. eth	Cx. van	Cx. ann	Cx. poi	Ere. spp	Fi. spp	Hod. spp	Ort. spp	Ura. spp	
High-land	Borowonin	0	3	0	2	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0	0	0	9
	Kapkenda	62	0	0	13	0	0	0	0	16	0	0	0	0	1	0	0	1	1	0	0	0	94
	Kaptimbor	128	2	0	24	1	0	0	0	0	0	0	12	4	4	0	0	0	1	2	0	0	178
	Kaptich	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Kiberege	6	0	2	0	0	0	0	0	2	0	0	0	0	0	0	0	0	1	0	0	0	11
	Talai	2	0	0	0	0	0	0	0	3	0	0	0	0	0	0	0	0	1	0	0	0	6
Subtotal		198	5	2	39	1	0	0	0	25	0	0	12	4	5	0	0	1	4	2	0	0	298
Spp %		66	1.7	0.7	13.1	0.3	0	0	0	8.4	0	0	4.0	1.3	1.7	0	0	0.3	1.3	0.7	0	0	
Low-land	Kapkuikui	2945	19	2	53	0	0	25	6	3197	74	1	0	1	0	0	0	0	168	0	0	2	6493
	Loboi	382	50	18	127	77	0	137	3	616	14	4	0	22	0	0	0	0	67	0	0	8	1525
	Nteppes	190	28	11	17	1	0	0	0	209	17	0	0	0	0	0	0	0	0	0	1	0	474
	Robert's	15	8	1	1	0	0	0	0	7	0	0	0	20	0	0	0	0	5	0	0	0	57
	Salabani	17	1	2	25	0	0	0	0	105	113	0	2	0	0	2	0	0	0	0	0	0	267
	Sirata	103	61	263	135	0	0	134	3	160	59	31	0	0	0	0	1	0	0	0	0	0	950
Subtotal		3652	167	297	358	78	0	296	12	4294	277	36	2	43	0	2	1	0	240	0	1	10	9766
Spp %		37.4	1.7	3.0	3.7	0.8	0	3.0	0.1	44.0	2.8	0.4	0.0	0.4	0	0.0	0.0	0	2.5	0	0.0	0.1	
Mid-land	Chebarsiat	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	Kimalel	0	0	0	4	0	0	5	0	6	0	2	0	0	0	0	0	0	0	0	0	0	17
	Kimao	1	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	3
	Kipcherere	3	1	3	1	0	0	1	0	16	0	0	0	0	0	0	0	0	1	0	0	0	26
	Kabeswa	0	0	0	2	0	0	0	0	12	0	0	0	0	0	0	0	0	0	0	0	0	14
	Yomu	0	0	0	1	1	0	1	0	5	0	0	0	2	0	0	0	0	0	0	0	0	10
Subtotal		4	2	3	8	1	0	7	0	40	0	2	0	2	0	0	0	1	0	0	0	70	
Spp %		5.7	2.9	4.3	11.4	1.4	0	10	0	57.1	0	2.9	0	2.9	0	0	0	1.4	0	0	0		
Riverine	Barwessa R	88	27	26	38	1	0	1	1	4	3	0	0	0	1	0	1	36	0	00	0	227	
	Barwessa S	2	7	3	1	5	0	0	0	27	0	0	0	0	0	0	0	14	0	0	0	59	
	Enot	13	1	6	51	0	4	3	0	66	0	1	0	0	0	0	0	0	0	0	2	0	147
	Kamnarok	12	64	60	43	0	142	14	55	912	11	0	0	0	0	0	2	0	12	0	0	0	1327
	Litein	16	5	1	4	0	0	4	1	28	0	0	0	0	0	0	0	0	0	0	29	0	88
	Salawa	1	5	1	0	0	0	0	0	16	0	0	0	0	0	0	0	0	3	0	0	0	26
Subtotal		132	109	97	137	6	146	22	57	1053	14	1	0	0	1	2	1	65	0	31	0	1874	
Spp %		7.0	5.8	5.2	7.3	0.3	7.8	1.2	3.0	56.2	0.7	0.1	0	0	0	0.1	0.1	0.1	3.5	0	1.6	0	

*Aedes species included: *Ae. hirsutus*, *Ae. vittatus*, *Ae. metallicus*, *Ae. aegypti* and *Ae. tarsalis*; Spp % - Species percentage; Cx-Culex; pip-pipiens; uni-univittatus; Ma-Mansonia; afr-africana; unif-uniformis; Ae-Aedes; Spp-species; Aed-Aedeomyia; A.n-Anopheles; cou-coustani; fun-funestus; gam-gambiae; pha-pharoensis; Coq-Coquillettidia; mac-macfieii; eth-ethiopicus; van; vansomereni; ann-annulioris; poi-poicilipes; Ere-Eretmapodite; Fi-Ficalbia; Hod-Hodgesia; Ort-Orthopodomyia; Ura-Uranotaenia.

Though RVF vector species were represented in all zones irrespective of sites, the distributional abundance varied for species in each zone. For *Mansonia africana*, 74.8%, 23.9%, 0.8% and 0.5% were collected at lowland; riverine, midland and highland zones respectively while *Ma. uniformis* was also collected in the lowland, riverine, midland and highland in the following proportions respectively: 66.1%, 25.2%, 1.5% and 7.2%. Proportions of *Cx. pipiens* s.l. collected were 91.6% in the lowland, 3.3% in the riverine, 0.1% in the midland and

5.0% in the highland while those for *Culex univittatus* were 59.0%, 38.5%, 0.7%, 1.8% respectively. The abundance of *Mansonia* species combined was significantly different in lowland compared to highland which was used as a reference ($p=0.018$). Similarly, abundance of *Cx. pipiens* s.l. was significantly different in the lowland compared to highland ($p=0.000$). However, *Culex univittatus* abundance was not statistically different across zones ($p>0.05$) (Table 4).

Table 4: Negative binomial coefficients for vector abundance and distribution across ecological zones

Zone	Species	Estimate	Std Error	z-value	p-value
Lowland	<i>Mansonia</i> spp	1.1860	0.05014	2.385	0.018
	<i>Cx.pipiens</i>	2.5730	0.3662	7.026	0.000
	<i>Cx.univittatus</i>	0.8695	0.7210	1.206	0.228
Midland	<i>Mansonia</i>	-1.0885	0.7363	-1.478	0.139
	<i>Cx.pipiens</i>	-1.7047	1.0040	-1.698	0.089
	<i>Cx.univittatus</i>	-0.9163	1.1339	-0.808	0.419
Riverine	<i>Mansonia</i>	0.6038	0.5245	1.151	0.249
	<i>Cx.pipiens</i>	-0.5108	0.3946	-1.295	0.195
	<i>Cx.univittatus</i>	0.5170	0.7243	0.714	0.475

Presence of RVF potential vectors in all ecological zones though in small numbers may point out to expansion of areas at risk. This is in agreement with the findings of Ochieng *et al.* which predict that future climatic conditions will lead to increase in spatial distribution of RVF vectors [28]. *Mansonia* species which are the principal vectors of RVF in Baringo [14], were collected in relatively large numbers from sites which were near swamps in each ecological zone. These sites included Kapkenda and Kaptimbor in the highlands; Barwessa River, Enot and Kamnarok in the riverine; Lobo and Sirata in the lowlands. In particular, Lobo and Sirata have large permanent swamps with the latter having the largest number of *Mansonia* mosquitoes collected in this study and was one of the hotspots of RVF during the 2006/2007 outbreak in Baringo. The large number of *Mansonia* species collected from sites near a swamp is similar with findings of Arum *et al.* in which more mosquitoes of this genus were collected from marshy areas [24]. An entomological survey in diverse regions of Kenya also indicated that *Mansonia* species are adapted to large swampy areas around lakes which provide suitable breeding habitats [15].

4. Conclusion

This study demonstrates the local abundance and distribution of mosquito species that could propagate arboviral infections in Baringo; information which is an integral component of risk assessment for localized RVF outbreaks. High densities of RVF vector mosquitoes are mostly concentrated in the lowlands where RVF outbreak occurred in 2006/2007 and riverine zone where RVF outbreak has never occurred. Presence of known vectors in areas previously free of RVF (riverine zone) is a risk indicator for future outbreaks. This raises concern as consequences of RVF outbreak are dramatic both for human and animal health. These findings should be incorporated in the existing national RVF contingency plan for improved local preparedness and early response to RVF outbreaks. Based on this study, frequent monitoring of mosquito vector species, distribution and abundance to detect geographical expansion of RVF vectors is recommended as one of the useful early warning signs for the local occurrence

and transmission of Rift Valley fever virus. The RVF vector distribution predictive map drawn from data generated in this study [28] will complement intervention measures for control of the disease, including strategic vaccination for livestock. Furthermore, the area –specific vector predictive maps can be incorporated into the existing national RVF contingency Plan by the Baringo Veterinary department as part of an improved local preparedness and early response in case of RVF outbreaks.

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