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Epidemiological patterns of Rift Valley Fever from diverse habitats during an extreme unprecedented flooding of Lake Baringo basin, Kenya, 2012-2013

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Abstract

Mosquitoes' ecology and associated arboviruses are heavily influenced by precipitation and retention of water in the environment. In 2011 and 2014, unprecedented floods occurred in Lake Baringo basin inundating approximate 88 km2 of the shoreline land. This caused abrupt environmental changes raising fears of an outbreak of Rift Valley Fever (RVF) disease. This study was carried out to determine the situation of RVF disease in livestock from diverse habitats during the extreme unprecedented flooding phenomenon that occurred in Lake Baringo basin, in 2012-2013. Blood was drawn from ear vein of livestock selected randomly from the three study areas (lakeshore land, swamp marshy and dry rangeland habitats). Mosquitoes were trapped using CDC light traps and identified morphologically. From a total of 77 blood samples, eight were positive for RVF virus (RVFV) representing an overall infection of 12%. RVF prevalence from livestock resident in flooded lakeshore land habitat was 2.6% (N=77) compared to the swamp marshy habitat at 7.8% (N=77). No infections were recorded from dry rangeland (0%). Mosquitoes of genus Mansonia dominated the catches in flooded lakeshore (98%). Highest individual catches of mosquitoes of genus Aedes was from swamp marshy area whose abundance was 96.8% and below 2% in other habitats. The Simpson's Diversity Index for mosquitoes from swamp marshy habitat was 0.56, dry rangeland 0.57 and lakeshore land 0.13. The flooded lakeshore land was the most affected by the unprecedented floods resulting in uneven mosquito diversity and subsequently low prevalence of RVF in this habitat. This could be attributed to prolonged disruption of biotic and abiotic factors creating unfavourable breeding sites of multiple species of primary vectors of RVF in flooded lakeshore land unlike in other habitats.

Keywords: *Lake Baringo; unprecedented floods; RVF prevalence; livestock; habitat*

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Introduction

Stratification of an ecosystem into several diverse habitats is important in understanding environmental connection with the hosts, pathogens and vectors (Restrepo *et al*., 2016). These variations help in giving an insight into spatial epidemiological patterns of a disease that could be a precursor to an outbreak (Wangara *et al*., 2019). Lake Baringo basin harbours diversity of habitats whose composition of fauna and flora is influenced by human activities and environmental changes (Odada *et al*., 2006). The basin has

severally experienced outbreaks of Rift Valley Fever (RVF) which is a zoonotic disease attributed to mosquitoes and floods caused by El nino rains (Nanyingi *et al*., 2015; Tigoi *et al*., 2015).

The RVF virus belongs to order Bunyavirales, Family Phenuiviridae and genus *Phlebovirus* (Abudurexiti *et al*., 2019). The disease was first described in Kenya in 1931 (Daubney *et al*., 1931). The Rift Valley Fever virus (RVFV) is able to invade diverse ecological systems (Jost *et al*., 2010). During heavy rains and particularly El nino that cause floods, there are increased number of breeding sites for the mosquitoes and hence intensifying virus transmission and circulation (Anyamba *et al*., 2001). The disease primarily affects cattle, sheep, and goats, but also people (Jost et al., 2010). Export and import of infected livestock is believed to be responsible for transmission across the borders of different countries (Chevalier *et al*., 2004). One of the most effective methods of detecting arboviruses is by use of Polymerase Chain Reaction (PCR) (Johnson *et al*., 2012).

The RVF disease epidemics usually occur after every 10 years with the virus able to invade diverse ecological systems (Jost *et al*., 2010). In South Africa, the disease was reported first in 1950, Namibia in 1955 and Egypt in 1977-1978. In 2000, an outbreak in Saudi Arabia caused death of 224 people (Chevalier *et al*., 2004). Kenya have experienced 11 national outbreaks of RVF with an average inter-epizootic period of three to four years between 1951 and 2007 (Nanyingi *et al*., 2015). The outbreaks were mostly attributed to Elnino/Southern Oscillation phenomenon (ENSO) rains causing flooding (Nanyingi *et al*., 2015) with the notable incidents having occurred in 1997-1998 (Okech *et al*., 2019) and 2006-2007 (Sang *et al*., 2010). In 2006/2007, an outbreak was reported in Garissa, Ijara, Maragwa, Thika, Baringo and Kilifi (Breiman *et al*., 2008; Munyua *et al*., 2010). The outbreak had the highest livestock morbidity and mortality rates including 90 human deaths (Breiman *et al*., 2010; Nguku *et al*., 2010). On a macroeconomic scale, losses from an outbreak of RVF in 2006-2007 is estimated to have been in range of Ksh 2.1 billion (US$32 million) on the Kenyan economy, based on its negative impacts on agriculture and other sectors like transport and services among others (Rich & Wanyoike, 2010).

Flooding, either from rainfall, artificially induced or unprecedented in any form results in creation of several ecotope layers in a habitat that affect mosquitos' distribution, breeding and eventual diseases transmission (Paula *et al*., 2012). For a period of over 30 years, Lake Baringo was experiencing a decreasing trend of the waters, from a depth of 8m in 1976 to 1.7m in 2001 (Okech *et al*., 2019). The area covered by water along the shores of the lake followed a similar trend with submerged land shrinking from 219km^2 in 1976, 136km^2 in 1986, to 114km^2 in 2001 (Okech *et al*., 2019). In 2010 to 2014, unprecedented and extreme floods occurred in Lake Baringo as a result of the rise in lake waters that displaced people from their homes and submerged acres of land along the shores. Gradual overflow of waters into the land around the lake covered expansive areas, soaking the vegetation, animal and domestic wastes. This was a rare form of flooding phenomenon said to recur after every 50 years and caused by geological disturbances and earths' tectonic movements (Obando *et al*., 2016; Okech *et al*., 2019). According to Onywere (2013b), between January 2010 – September 2013, the floods resulted in water rising from a low of 143.6 km^2 in January 2010 to a high of 231.6 km^2 in September 2013. The rising waters inundated 88 km^2 (61.3%) of lakeshore land surpassing any previous flooded water marks on the land (Obando *et al*., 2016).

This study aimed at assessing spatial risks, vectors and RVF prevalence in livestock resident in diverse habitats of Lake Baringo basin during the period of prolonged, extreme and unprecedented flooding in 2012-2013. The study identifies some of the critical environmental factors and exposure, the interaction with vectors, hosts and pathogens in sustenance of RVFV. The findings would be helpful in advising on vector policy, reviewing

of RVF disease risk maps and contingency plans.

Materials and methods

Study area

Lake Baringo basin is located in Baringo County, at an altitude of 950m above sea level, latitudes 00^{0}28'N and 00°32'N and longitudes 36^{0}58'60E and 36^{0}00'E. The lake and the basin is located in semi-arid area, ecological zone VI (Johansson and Svensson, 2002). It experiences bimodal rainfall with the long rain season starting from March–June and a short rain season from October – November, averaging 1200 mm per year with a range of 300-700mm (Wetang'ula *et al*., 2012).

Selection of study sites

Lake Baringo basin is regarded as an RVF endemic area (Tigoi *et al*., 2015). In the basin, three study sites were selected to represent three strata of habitats; flooded lakeshore land (represented by Ngambo village), swamp marshy (represented by Kapkuikui village) and dry rangeland habitat (represented by Kimelel) (Figure 1). The flooded lakeshore land included several villages with Ngambo selected to represent the rest because of its accessibility. Ngambo Village lies at latitude 0°30'30.6"N, longitude 36°03'36.7"E and an altitude of 995 metres above sea level. The area was used for subsistence crop and livestock farming. *Prosopis juliflora* was the dominant plant covering large tracks of land. Floating macrophytes covered most of water surface following flooding in the lakeshore ecological habitat. This type of vegetation is said to offer conducive habitat for mosquitoes breeding (Mwangi & Swallow, 2005).

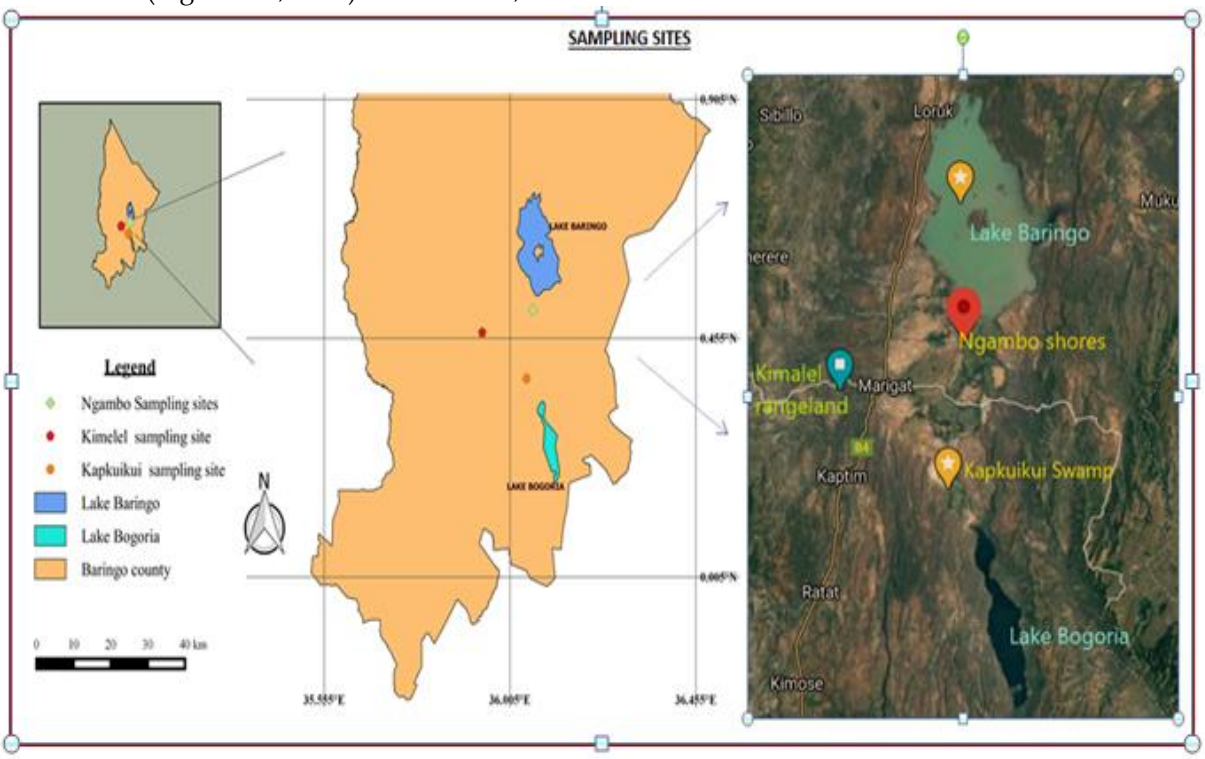

Figure 1. Location of study sites from Lake Baringo basin (flooding lakeshore land at Ngambo; swamp marshy at Kapkuikui and dry rangeland ecological habitats at Kimelel

The swamp marshy habitat located at Kapkuikui lies between Lake Baringo and Lake Bogoria at latitude 0°22'44.6"N, longitude 36°02'42.1"E and altitude of 1004 metres above sea level. It forms an extensive soggy ground of Loboi wetlands that are fed by underground and surface springs including River Loboi. The main features of the swamp are extensive

growth of emergent macrophytes dominated by *Cyperus papyrus* and *Typha domingenis* (Terer *et al*., 2012). It is a fragile ecosystem which is easily destabilized by both natural and anthropogenic activities (Odada *et al*., 2006). The area is extensively used as a communal grazing area with wildlife such as Zebras, Ostriches, and different species of antelopes flocking along with grazing livestock.

The dry rangeland habitat is located in Kimelel at latitude 0°27'57.2"N, longitude 35°56'15.7"E and altitude 1172 meters above sea level. There is a seasonal dam that fills with water during the rainy season but dries up during the dry season. The habitat is characterized by scattered acacia species, grass and sparse shrubs. Unlike the other habitats around Lake Baringo, only a few *P. juliflora* grow in the area. This habitat is separated from the other two by a sharp escarpment.

Sample collection

This was a longitudinal research carried out from October 2012 - October 2013. From the three study habitats, stratified random sampling of the homesteads was carried out where those rearing the four species of livestock; indigenous cattle, sheep, goats and poultry were profiled. To ensure there was no inter-effects of one homestead to the other, the selected homesteads stood at an approximate distance of one kilometre from each other. Each study habitat had two homes selected where livestock were sampled at the end of every two months. Whole blood was drawn from the ear vein but in case of indigenous poultry, blood was obtained from wing vein. Bar-coded vacutainer tubes coated with EDTA were used in both cases. The sample blood was aliquoted into 2mL cryovials, carried to laboratory in a container packed with iced CO_2 ready for storage and preservation in biorepository nitrogen liquid tanks awaiting further analysis. Monthly trapping of mosquitoes was carried out from the three habitats using CDC traps for 12 months. The catches from each habitat were identified using morphological features and recorded.

Sample size

The selection of livestock to be sampled in the research was purposive. Those aged above three years and had no clinical history of a disease over the last four months were recruited into the study. At the beginning of each sampling session, physical examination for any form of illness was done to the target animals by a veterinarian. Only those perceived to be ill and of poor body condition had their samples taken and eventually screened for RVFV. A total of 77 samples were screened using PCR of which 22 were cattle, 16 sheep, 17 goats and 22 poultry.

Identification of arboviruses

The procedure for Nested PCR was used to synthesise cDNA. The RNA was extracted from the livestock blood samples using QIAamp viral RNA extraction protocol (QIAQEN, Thermo Fisher, California, USA) to make total RNA. A volume of 200 µL sample blood was added to 60 µL of red blood cell lysis buffer, incubated for five (5) minutes at room temperature and then centrifuged for one (1) minute. The products were pre-filtered, isolated, purified and concentrated into a total RNA end product. The extracted RNA was then eluted with ≥6 µL of RNase-free water. The First cDNA was synthesised from extracted total RNA using Superscript Kit II (Invitrogen, Thermo Fisher, California, USA) protocol and then stored in a freezer at -20^{0}C. The Second Strand cDNA was synthesized as described in the QIAGEN quick PCR Purification Kit (QIAGEN, Thermo Fisher, Hilden, Germany) in readiness for amplification. Sequences of the primer used in the study (5′-ATG-CTG-GGA-AGT-GAT-GAG-CG-3′) and (5′-GAT-TTG-CAG-AGT-GGT-CGT-C-3′) were as defined in Terrestrial Manual for OIE (OIE Terrestrial Manual, 2016).

Statistical analysis

The data collected was analysed using SPSS software Ver.16 and Microsoft Office Excel Data Analysis Ver. 2013. Simpson's Diversity Index was calculated using an online diversity calculator (Young, 2021). The percentage of

livestock species infected with RVF disease was expressed as a period prevalence. The significance differences in infection for livestock species resident in the three ecological habitats was calculated using ANOVA at $p<0.05$. For paired comparison between two habitats, t-test was set at $P<0.05$. Odds ratio (OR) was used to compute the measure of association for the disease and exposure to dominant vector in flooded shoreline.

Results

Mosquito infestation in the three habitats from Lake Baringo basin

A total of 386,625 individual mosquitoes were trapped in the three study habitats. The flooded lakeshore land habitat had the highest catch of mosquitoes constituting 88.9%, swamp marshy 10.8% and dry rangeland 0.3%. Eleven genera were caught; *Anopheles*, *Mansonia, Coquillettidia, Culex, Ficalbia, Aedomyia, Aedes, Theobaldia, Uranotaenia, Orthopodomyia,* and *Hodgesia*.

Mosquitoes of genus *Mansonia* (constituting *Mansonia africana* and *M. uniformis*) dominated flooded lakeshore land habitat with a catch of 98%. The swamp marshy area was the preferred habitat for three genera of mosquitoes that are key in transmission of RVF; *Aedes*, *Culex* (constituting *Culex pipens pipens*, *Culex pipens quinquefasciatus* and *Culex tritaeniorhynchus* species) and *Coquillettidia* with an abundance catch of 96.9%, 64.1% and 53% respectively. In the dry rangeland area, the catch of *Aedes* mosquitoes was 1.2%, *Culex* 0.5% and *Mansonia* below 0.1%. Flooded lakeshore land habitat had the lowest mosquitoes' Simpson's Diversity Index of 0.13, while the swamp marshy and dry rangeland had Simpson's Diversity Index of 0.56 and 0.57 respectively.

Prevalence of RVF infection

The overall prevalence of the RVF disease in livestock species from the basin during the period of extreme unprecedented flooding of Lake Baringo was 12% (N=77). Sheep had the highest risk of infection at 19% prevalence, followed by Goats with 12% and Cattle at 9%. Poultry had the lowest prevalence of infection for RVF at 5%.

Prevalence of RVF infection in diverse habitats

The prevalence and potential risk to infection differed with the type of habitat. The highest risk of livestock to infection with RVF disease was from the swamp marshy habitat at 7.8%, followed by flooded lakeshore land at 2.6%. Livestock from the dry rangeland were not infected (Table 1). The measure of association computed as odds ratio (OR) showed that a statistical association existed between the disease and exposure to mosquitoes of genus *Mansonia* in flooded shoreline. The odds of infection to livestock resident in the flooded shoreline habitat that was heavily infested with *Mansonia* (98%) was 8.0 times higher than in dry rangeland habitat where infestation was below 0.1%.

From the study, goats from the swamp marshy habitat faced the highest risk to the disease with RVF prevalence of 40%, followed by sheep at 25%. In the flooded lakeshore land, sheep was the most affected with RVF prevalence of 33%. In this study, it emerged that the habitat had a significant role at $p<0.05$ in transmission of RVF disease ($F_{(2, 9)} = 10.5$ $p=0.004$). The Tukey HSD test for multiple comparison showed a significance difference in infections between livestock resident in swamp and flooded habitats ($p<0.036$); between swamp and dry rangeland ($p<0.003$). However, there was no significant differences in infection based simply on the species of livestock ($F_{(3, 4)} = 0.02$ $p=0.5$).

Table 1. Risk of livestock to infection with RVFV from the three ecological habitats of Lake Baringo basin, Kenya

	Swamp marshy habitat		Flooded habitat		Dry rangeland habitat		Total	
Livestock species	**n**	**+ve**	**n**	**+ve**	**n**	**+ve**	**N**	**+ve**
Goats	n=5	2 (40%)	n=5	0 (0%)	n=7	0 (0%)	N=17	2 (12%)
Sheep	n=8	2 (25%)	n=3	1 (33%)	n=5	0 (0%)	N=16	3 (19%)
poultry	n=8	1 (13%)	n=8	0 (0%)	n=6	0 (0%)	N=22	1 (5%)
Cattle	n=8	1 (13%)	n=6	1 (17%)	n=8	0 (0%)	N=22	2 (9%)
(+ve/n)	**n=29**	**6 (21%)**	**n=22**	**2 (9%)**		**0 (0%**		
(+ve/N)		**7.8%**		**2.6%**		**0%**	**N=77**	**8 (12%)**

Discussion

Studies of an ecosystem in respect to environment, ecology, interaction of the vector with diverse hosts and other demographic variations help in mapping and description of risk factors including understanding spatial epidemiological patterns of an infectious disease (Elliot & Wartenberg, 2004). In this study, the interaction of the changing environmental factors with multiple species of the mosquitoes that vector RVF and hosts from the habitats facilitated circulation and sustenance of the virus in Lake Baringo basin. Previous studies have associated outbreaks of RVF disease with El nino rains and ensuing floods. The major outbreak of 2006/2007 in the Lake Baringo was attributed to El nino rains that covered the entire basin (Breiman *et al*., 2010; Nguku *et al*., 2010).

In our study, the lakeshore habitat was the most affected by the unprecedented extreme floods of 2012-2013. The rising lake waters never reached the swamp marshy and dry rangeland habitats. Normal floods associated to seasonal rains were not experienced during the study period in the basin. The low risk of infection with RVF encountered from the lakeshore land habitat was as a result of interaction of the altered environmental factors within the ecology occasioned by excess lake waters. The lake waters flooding lakeshore land habitat, emergence of floating macrophytes favoured breeding and proliferation of *Mansonia africana* and *M. uniformis* species of mosquitoes to other genera giving rise to uneven mosquito diversity. Dense *P. juliflora* protected them from desiccation. Decomposing submerged animal waste and vegetation including other debris modified the biological, environmental and physical factors within the habitat and breeding sites of mosquitoes that are primary vectors of RVF disease.

The swamp marshy habitat supported diverse mosquitos' genera with the high risk of infection with RVFV exacerbated by high catches of genera *Aedes* and *Culex* species that included *Culex pipens pipens*, *Culex pipens quinquefasciatus* and *Culex tritaeniorhynchus* species. According to Seufi (2010), these species acts as primary and secondary vectors of RVF. Fresh water from the springs, broad and axial leaved plants, and occasional precipitation including scattered pools of water favoured breeding and survival of diverse species of mosquitoes that vector RVFV.

Studies by Lichoti (2014) in Lake Baringo basin showed that in times of normal rains, livestock resident in the lakeshore land area were at highest risk of infection with RVFV than in other habitats with the risk averaging 26.5% - 24.4%. Similar findings were documented in a study by Munyua (2010) during El nino rainy

season experienced in 2006/2007 with the RVF prevalence ranging from 36.5% -27.8%. Interestingly, the study by Munyua (2010) recorded 0% infections in livestock from swamp marshy area (Kapkuikui village) whereas our study recorded a 21% infection of those resident in the habitat. This study shows a new epidemiological pattern of exposure to mosquitos' of genus *Mansonia* and the RVF disease in flooded shoreline. In this habitat, the odds ratio of 8.0 meant that the circulation and sustenance of the RVFV in livestock can be attributed to heavy habitation by mosquitos' of genus *Mansonia*. This highlights the importance of spatial epidemiology and stratification of an ecosystem in understanding prevailing risk factors that aid in sustenance and outbreak of RVF.

Poultry resident in swamp marshy and flooded lakeshore land area were shown to be infected with RVF virus. The two habitats harboured the mosquitoes of genus *Coquillettidia* which according to Molaei (2008) is mostly ornithophilic but can feed on other available domestic animals and humans. The study by Molaei (2008) further clarified that the genus supports enzootic amplification of RVFV and facilitate transmission from non-viremic birds to mammals. In this study, it is therefore high likely that the non-viremic RVFV in poultry could have been one of the sources of circulating virus in the Lake Baringo basin.

Conclusion

In conclusion, unlike the floods caused by heavy rainfall, the rise of the waters of Lake Baringo basin during the extreme unprecedented flooding in 2010-2014 was limited to expansive lakeshore land habitat. The flooding lake waters caused prolonged disruption of biotic and abiotic factors creating unfavourable breeding sites for most of mosquitos' species and subsequently low prevalence of RVF. Exposure of livestock to mosquitoes of genus *Mansonia* ensures circulation of RVFV at low levels in a herd which can be a source of an outbreak in case of habitation by other primary vectors under and existence of favourable environmental conditions. Flocking of livestock and wildlife together, exposure to diverse species of mosquitoes and existence of numerous small scale scattered microhabitats optimal for mosquitoes breeding are key primary factors in sustenance of RVF in livestock in swampy marshy habitat. With adequate precipitation, the swamp marshy ecology is a potential source of an outbreak in an endemic RVF ecosystem.

Recommendations

This study recommends timely vaccination of livestock resident in Lake Baringo basin against the RVF disease and establishment of sentinel livestock herd from the area for continuous surveillance of the disease. There is a need for further study to understand the transmission of non-viremic RVF to domestic poultry and the role they play in sustenance of the RVF virus in livestock

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