Risk Factor Analysis for Outbreak of Rift Valley Fever in Khartoum State of Sudan[1]

Deena M. Abdelgadir,[2] Hind M.M. Bashab, Rania A. Elhadi Mohamed, and Sara A. Abuelmaali

University of Khartoum, Sudan

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

J. Entomol. Sci. 45(3): 239-251 (July 2010)

Abstract Baseline surveys were conducted in Khartoum State, Sudan, during the rainy season (end of July to the beginning of September) in 2008, and female mosquitoes were sampled for arboviruses from November 2008 to January 2009 to predict outbreaks of Rift Valley Fever (RVF) in Khartoum State. *Aedes vittatus* (Bigot) and *Ae. vexans* (Meigen) vectors were found only at one of the study sites (Soba West). Under laboratory conditions, survival of these mosquitoes exceeded the mean incubation period of the arboviruses in these vectors (15 days). Field-collected blood-fed female mosquitoes were tested for the RVF virus using the real-time PCR technique. The virus was detected in populations of *Ae. vexans* and *Culex quinquefasciatus* (Say) for the first time in Sudan.

Key Words Rift Valley Fever Virus (RVFV), survival, *Aedes vexans, Culex quinquefasciatus*

Arthropod-borne viruses (arboviruses) comprise the largest class of vector-borne human pathogens. Many species of mosquitoes transmit arboviral diseases which afflict man and his animals. These include dengue fever/dengue hemorrhagic fever (DF/DHF), yellow fever, Rift Valley Fever (RVF), and the West Nile Viruses (WNV) (Gubler 1998, Jupp 2005).

Rift Valley Fever is caused by a virus belonging to Bunyaviridae. It is a zoonotic disease and can be transmitted from animals to humans (Gubler 2002). RVF is a significant threat to human health in endemic countries of Africa and the Middle East. The disease also results in significant economic losses due to death and abortion among RVF-infected livestock.

The disease is emerging and resurging as a health problem due to climatic as well as societal changes. In recent years, devastating outbreaks of the disease have been reported from several countries (Gubler 2002). The first RVF outbreak in Sudan was recorded in 1973 in the Kusti area; a second occurred in 2003. During the second outbreak, suspected cases were recorded in Khartoum State (FMOH 2008). The most recent outbreak of RVF occurred between 2007 and 2008. During this outbreak, the recorded number of RVF human cases in Sudan reached 566 in 2007 and 698 in 2008 with 222 and 178 deaths reported, respectively (WHO 2007a, 2008).

Female mosquitoes of the genera *Culex* and *Aedes* transmit the RVF virus (e.g., *Ae. aegypti*, *Ae.vexans* and *Ae.ochraceaus*) (Favier et al. 2006). RVF can also be

[1]Received 03 August 2009; accepted for publication 03 January 2010. The authors have made equal contributions to the completion of this research and manuscript.

[2]Address inquiries (email: deenagader@yahoo.com).

transmitted by many different species that have a global distribution (Gubler 2002). It also is transovarially transmitted. This accounts for the continued presence of this virus in foci and provides the virus with a survival mechanism as the eggs of these mosquitoes can survive for several years in dry conditions. During periods of heavy rainfall, larval habitats frequently become flooded enabling infected eggs to hatch and the resultant mosquito populations to rapidly increase and, thus, transmitting the virus to the animals from which they take blood meals (Wilson 1994). Unfortunately, failure of containment of outbreaks of this disease is mainly attributed to inadequate knowledge of the ecology of nonmalaria mosquitoes.

Whereas most human cases of the disease are relatively mild, a small percentage of patients develop a much more severe form of the disease. Neither effective treatments nor vaccines are available, thus, vector control is considered to be the most effective method for the prevention and control of this disease (Gubler 1998). Forecasting can predict climatic conditions that are frequently associated with an increased risk of outbreaks and may improve disease control. In Africa, Saudi Arabia and Yemen, RVF outbreaks are closely associated with periods of above-average rainfall (Barry et al. 2002). Within the framework of the 2005 International Health Regulations, the forecasting and early detection of RVF outbreaks, together with a comprehensive assessment of the risk of spread to new areas, are essential for effective and rapid control measures to be implemented (WHO 2007b). The risk of arbovirus transmission varies depending on the immunity, density of the host population, mosquito-host interactions, route of transmission, and both biotic and abiotic factors, such as temperature, relative humidity, mosquito susceptibility to viruses which affect the biology of mosquitoes as well as mosquito-virus interactions and, subsequently, the risk of outbreaks of hemorrhagic fevers (Scott and Morrison 2003).

This research aimed to forecast the risk of RVF outbreak in Khartoum State through screening field-collected blood fed female mosquitoes for the presence of the virus as well as investigating their survival longer than the 15-d incubation period of the RVF virus in mosquitoes.

Materials and Methods

The State of Khartoum is located between 15 10" and 16 30"N latitude and 31 35" and 40 20"E longitude. Six study sites were chosen depending upon certain characteristics. The Khartoum Bus Terminal (Site A) was selected because of the risk of passive transport of viruses as well as infected mosquitoes by buses and travelers arriving daily from different locations in Sudan. Site B was the Khartoum North Central Livestock Market where sheep from different parts of Sudan arrive. Shambat (Site C) and Hellat KuKu (Site D) were selected due to the occurrence of suspected cases of RVF during the last few years (FMOH 2008). Hellat KuKu also was selected due to the presence of nomadic tribes. Soba West (Site E) was selected due to the presence of *Ae.vexans* mosquitoes. Al Shegelab (Site F) and Omdurman (Site G) also were used as study sites.

Baseline cross-sectional surveys were conducted during July 2008 through January 2009. Seventy-five surveys were conducted at 5 sites (C, D, E, F, G) for estimating mosquito survival rates; 70 surveys were conducted at 4 sites (A, B, E, D) for the purpose of surveillance of arboviruses in blood-fed female mosquitoes.

For each survey event, 4th-instar larvae, pupae, and newly-emerged adult females were collected from each site, and reared in the laboratory to estimate survival rates.

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

A total of 6,436 blood-fed female mosquitoes was collected from the 4 study sites. The sample size of 1609 individuals was recommended by Weidong and Robert (2004) for high probability (0.8) of detection of the virus. Of the 1609 specimens that were randomly collected, 1424 were *Culex* sp. and 185 were *Aedes* sp.

Resting adult mosquitoes were collected from inside rooms and outdoor earthen jars 'zeers', septic tanks, and wells using aspirators made of glass tubing 29 cm long (diam 1.4 cm), One end of the tube was covered with fine wire screening and attached to a piece of rubber tubing 30 - 60 cm long. Mosquitoes were separated according to genera then placed into labeled cups covered with a piece of mosquito net and a rubber lid. A small slit was made at the middle of the net and covered with a piece of cotton to prevent mosquito escape during aspiration. Collected mosquitoes were used for the RVF virus surveillance studies.

Mosquito larvae were scooped from breeding sites using a 350-mL container. Pupae and 4th instars were transferred to white pans in the laboratory. The water of their breeding sites was used as a nutritive source. Pupae were collected and kept in 30x30x30 meshed cages in which they emerged after 2 d. The immature stages and adult mosquitoes were kept in a controlled environment at 27 ± 1°C, 85 ± 5% RH, and light-dark photophase of 12:12 h (Gad et al. 1987).

Newly-emerged mosquitoes were used to establish 3 cohorts of the 2 genera (*Culex* and *Aedes*) for each study site. Each cohort was maintained in a separate cage. Three cages per study site were randomly assigned to treatments of either: (1) starved mosquitoes, (2) allowed to feed on 10% sucrose solution (w/v), or (3) a combination of 10% sucrose and periodic pigeon blood meals. Blood-fed mosquitoes were offered blood meals every 2 d (Beier 1996). Dead mosquitoes were separated by gender, counted, recorded, and removed daily.

The field-collected blood-fed female mosquitoes that were assayed for the RVF virus were first identified to species using the keys of Hopkins (1952). Viral RNA was extracted from groups of the mosquitoes using a modified Jupp et al. (2000) method. Groups of mosquitoes were triturated with mortar and pestle in varying volumes of medium depending on the group size as follows: 10 - 50 mosquitoes (2.0 ml), 100 mosquitoes (3.0 ml), 150 mosquitoes (4.0 ml), 200 mosquitoes (5.0 ml), and 300 mosquitoes (8.0 ml). The medium was the Leibovitz medium with 10% fetal calf serum, antibiotics, and fungicide. The supernatants were filtered through a 3.5-µm filter and centrifuged at 3,000 cpm for 30 min. The supernatants were used for the RNA extraction. Viral RNA was extracted from clarified supernatant using a QIAamp viral RNA kit (Qiagen, Hilden, Germany) according to the instructions of the manufacturer.

The samples were first lysed by adding lyses buffer. The temperature was elevated so as to inactivate RNases and to insure isolation of intact viral RNA. Buffering conditions were then adjusted to provide optimum binding of the RNA to the QIAamp membrane, and the samples were loaded onto the QIAamp mini spin column. The salt and pH conditions insured that the RNA binded to the silica gel column during 2 centrifugations. The RNA bound to the membrane and contaminants were washed away in 2 steps using 2 different wash buffers. High-quality RNA was eluted in a special RNAse-free water. The purified RNA was free of protein, nucleases, and other contaminants and inhibitors. RNA was stored at -40°C until the RT-PCR was performed.

An RT-PCR was performed using the RT-PCR tube. Depending on the number of samples, the mixture was prepared as follows: 11.2 µL of the master mix, 0.8 µL of the mix enzyme (Taq polymerase and Reverse transcriptase) and 12 µL of the

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

Table 1. Longevity of field-collected *Culex* female mosquitoes fed on blood meals in the laboratory.

Days	% cumulative mortality of *Culex* mosquitoes				
	Site D (n = 67)	Site F (n = 19)	Site C (n = 26)	Site E (n = 16)	Site G (n = 32)
5	30%	47%	77%	44%	28%
10	46%	53%	85%	81%	28%
15	57%	58%	85%	88%	34%
20	61%	58%	85%	88%	59%
25	70%	58%	85%	88%	63%
30	79%	58%	92%	100%	63%
35	81%	63%	92%	-	72%
40	88%	63%	96%	-	81%
45	93%	74%	96%	-	94%
50	100%	89%	96%	-	97%
55	-	100%	100%	-	100%
r^2 (P = 0.05)	0.96	0.794	0.92	0.5929	0.96
r (P = 0.05)	0.98	0.891	0.96	0.77	0.981
Regression equation Y=	32.87 + 1.34X	39.8 + 0.86X	77.8 + 0.4X	66.6 + 0.78X	17.96 + 1.58X
The lower & upper limit of the 95% confidence interval for the slope	1.14 —— 1.55	0.53 —— 1.19	0.31 —— 0.5	0.29 —— 1.3	1.34—— 1.82
The lower & upper limit of the 95% confidence interval for the intercept	25.9—— 39.8	28.6 —— 50.98	74.74 —— 80.97	50.1——83.05	9.93 —— 25.99

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

template (extracted RNA). The Primer used was an Oligonucleotide specific primer with the following sequence:

NSca 5′-CCTTAACCTCTAATCAAC-3′ Map position: 841 - 824, orientation: Anti-sense.
NSng 5′-TATCATGGATTACTTTCC-3′ Map position: 31 - 48, orientation: Sense.

The PCR tubes were closed and transferred into the rotor of the RotorGene™ instrument. Denaturation occurred at 95°C (for 5 s), one step annealing and extension occurred at 57°C (for 35 s). The following steps were the cycling for amplification of cDNA (the total number of cycles was 45). Then, the sensitivity of the fluoresence channels was adjusted. Finally, the RotorGene ™ run was initiated.

Statistical analysis. Linear regression and correlations analysis were conducted with cumulative mortality (%) of female mosquitoes and days using NCSS 2007 and GESS 2006 software. Individuals that survived beyond the mean incubation period of the arboviruses within mosquitoes (15 days) were noted.

Results

Nutrition effects on adult mosquito mortality. The proportion of *Culex* females collected from the 5 sites (C, D, E, F, G), fed blood meals in the laboratory, and that survived longer than the 15-d incubation period for RVF virus in mosquitoes was 15% for Site C, 43% for Site D, 12% for Site E, 42% for Site F, and 66% for Site G (overall mean = 35.6%). Only 12% of *Aedes* females (Site E) survived for more than 15 d. Blood feeding was highly correlated with survival of female *Culex* and *Aedes* mosquitoes longer than the 15-d incubation period of the RVF virus in mosquitoes (Tables 1, 2).

Table 2. Longevity of field-collected *Aedes* female mosquitoes (Site E) fed on blood meals in the laboratory.

Days	% cumulative mortality of *Aedes* mosquitoes (n = 16)
5	38%
10	81%
15	88%
20	88%
25	100%
r^2 (P = 0.05)	0.75
r (P = 0.05)	0.87
Regression equation Y=	Y= 39.7 + 2.62 X
The lower & upper limit of the 95% confidence interval for the slope	−0.16 —— 5.399
The lower & upper limit of the 95% confidence interval for the intercept	−6.385 —— 85.785

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

Table 3. Longevity of field-collected female *Culex* mosquitoes fed on sugar solutions in the laboratory.

	% cumulative mortality of *Culex* mosquitoes				
Days	Site D (n = 51)	Site F (n = 46)	Site C (n = 34)	Site E (n = 9)	Site G (n = 55)
5	20%	11%	15%	67%	0%
10	24%	15%	15%	67%	0%
15	31%	17%	21%	78%	0%
20	33%	20%	24%	78%	49%
25	41%	22%	26%	89%	49%
30	45%	26%	29%	89%	49%
35	47%	33%	32%	100%	51%
40	51%	39%	38%	-	53%
45	53%	47%	44%	-	58%
50	59%	50%	47%	-	60%
55	61%	54%	50%	-	75%
60	67%	70%	53%	-	93%
65	73%	80%	59%	-	96%
70	82%	87%	68%	-	96%
75	84%	93%	71%	-	100%
80	88%	98%	74%	-	-
85	96%	100%	79%	-	-
90	98%	-	91%	-	-

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

Table 3. Continued

95	98%	-	94%	-	-
100	98%	-	100%	-	-
105	98%	-	-	-	-
110	98%	-	-	-	-
115	100%	-	-	-	-
r^2 ($P = 0.05$)	0.97	0.93	0.99	0.55	0.82
r ($P = 0.05$)	0.98	0.97	0.99	0.74	0.91
Regression equation Y=	−19.7 + 0.79X	4.099 + 0.99X	4.9 + 0.88 X	79.9 + 0.24X	14.8 + 0.93 X
The lower & upper limit of the 95% confidence interval for the slope	0.72 —— 0.86	0.87 —— 1.11	0.83 —— 0.93	0.14 —— 0.34	0.73 —— 1.13
The lower & upper limit of the 95% confidence interval for the intercept	15.3 —— 24.1	−4.12 —— 12.3	1.5 —— 8.3	73.18 —— 86.64	1.18 —— 28.5

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

Feeding of field-collected *Culex* females with sugar diets increased the respective proportions of those surviving longer than the 15-d incubation period with 79% at Site C, 69% at Site D, 22% at Site E, 83% at Site F, and 100% for Site G for *Culex* females (overall mean = 70.6%), whereas only 9% of *Aedes* females from Site E survived greater than 15 d. Sugar-feeding was highly correlated with the number of female *Culex* and *Aedes* mosquitoes that survived longer than the 15-d incubation period for RVF (Tables 3, 4).

Starved mosquitoes collected from the study sites did not survive beyond 5 d, far below the 15-d incubation period (Tables 5, 6).

Real-time PCR detection of RVF virus in mosquitoes. Each sample above 0.02 (the threshold) was considered positive for the virus, whereas samples below the 0.02 threshold were considered negative. Groups of *Ae.vexans* collected from Site E and *Cx. quinquefasciatus* collected from Site D exceeded 0.02 threshold and, thus, were considered to harbour the RVF virus (Fig. 1).

Discussion

Aedes vexans and *Ae. vittatus* produce drought-resistant eggs and, when infected with RVF virus, will provide a latent source of infection when females emerge after a rainfall event (Martin et al. 2008). Successful transmission of an arbovirus by a mosquito to the host requires sufficient time for the pathogen to develop in the mosquito and to be transmitted to a new host. Factors that affect mosquito survival during this

Table 4. Longevity of field-collected female *Aedes* mosquitoes (Site E) fed on sugar solutions in the laboratory.

Days	% cumulative mortality of *Aedes* mosquitoes (n = 11)
5	64%
10	91%
15	91%
20	91%
25	91%
30	91%
35	91%
40	100%
r^2 (P = 0.05)	0.49
r (P = 0.05)	0.7
Regression equation Y=	75.25 + 0.6 X
The lower & upper limit of the 95% confidence interval for the slope	–0.01 —— 1.21
The lower & upper limit of the 95% confidence interval for the intercept	59.85 —— 90.65

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

Table 5. Longevity of field-collected female *Culex* mosquitoes starved in the laboratory after collection.

Days	% cumulative mortality of *Culex* mosquitoes				
	Site D ($n = 55$)	Site F ($n = 49$)	Site C ($n = 39$)	Site E ($n = 18$)	Site G ($n = 63$)
1	55%	49%	67%	17%	67%
2	98%	65%	92%	61%	84%
3	100%	90%	95%	94%	84%
4	-	100%	95%	100%	100%
5	-	-	100%	-	-
r^2 ($P = 0.05$)	0.53	0.9	0.697	0.81	0.81
r ($P = 0.05$)	0.73	0.95	0.84	0.902	0.902
Regression equation Y=	63 + 9.2 X	39.7 + 13.74 X	69.1 + 69.1 X	–2.5 + 28.2 X	59 + 9.90 X
The lower & upper limit of the 95% confidence interval for the slope	–6.6 —— 25.01	1.07 —— 4.4	–1.45 —— 15.25	1.89 —— 54.5	–0.16 —— 19.96
The lower limit of the 95% confidence interval of the intercept	10.55 —— 115.4	12.0 —— 67.35	41.39 —— 96.81	–74.5 —— 69.5	31.4 —— 86.56

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

Table 6. Longevity of field-collected female *Aedes* mosquitoes (Site E) starved in the laboratory after collection.

Days	% cumulated mortality of *Aedes* mosquitoes (n = 23)
1	35%
2	52%
3	78%
4	100%
r^2 (P = 0.05)	0.994
r (P = 0.05)	0.997
Regression equation	Y = 11 + 22 X
The lower & upper limit of the 95% confidence interval for the slope	16.9 —— 27.32
The lower & upper limit of the 95% confidence interval for the intercept	–3.3 —— 25.29

period are critical to the onset and maintenance of epidemics of mosquito-borne disease and are important to understand because they provide specific targets for the action of mosquito control measures. Some species of mosquitoes can transmit the virus after 14 - 16 d; whereas, others transmit the virus after 6 d (Gad et al. 1987, Moutailler et al. 2007). Consequently, the effect of the sugar and blood meal under controlled laboratory conditions on the survival of female mosquitoes was evaluated in this study.

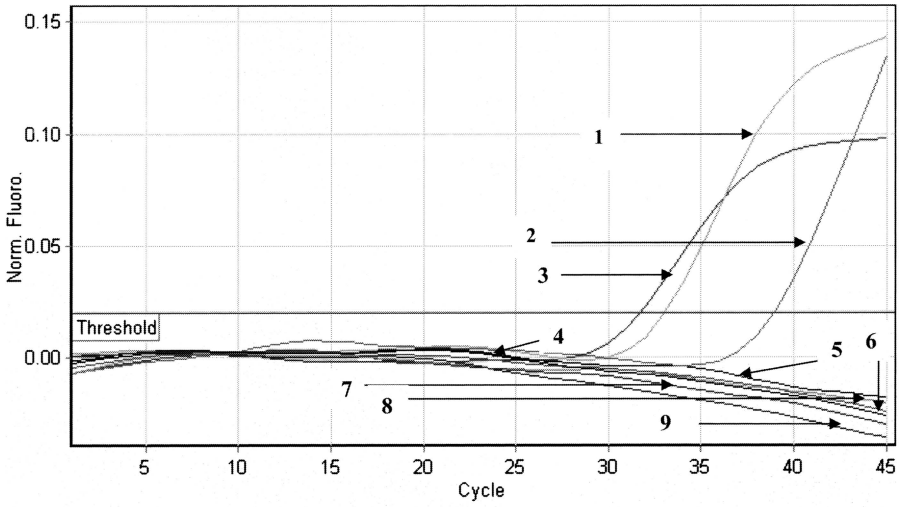
Results show that a significant proportion of females of the 2 genera survive longer than the incubation period of the viruses in the mosquitoes when fed either blood meals or sugar solution. This survival increases the risk of acquiring, maintaining and transmitting arboviruses. As much as the period of the survival is extended, the number of blood meals taken increases, thus increasing the probability of the transmission of viruses to large numbers of hosts is subsequently increased. Survival of mosquitoes for such a long period when fed on sugar indicates that the mosquitoes of the 2 genera can survive without blood meals for an extended period until they find appropriate host(s).

The mortality rates of females of mosquitoes were age-dependent, increasing with age. This corroborates a previous study of mortality and survival of wild populations of mosquitoes by Clements and Paterson (1981).

The effect of food type (sucrose 10%, sucrose 10% with blood) on the longevity of adult mosquitoes revealed an overall reduction in longevity of females maintained on a sucrose and blood diet compared with sucrose alone. These results corroborate findings of earlier studies performed with other species of mosquitoes (Nayar and Van Handel 1971, Nayer and Sauerman 1975a, b).

Assays for the virus were conducted only on blood-fed females because of the higher probability that these insects might harbor the virus. *Culex* and *Aedes* mosquitoes were given priority because they are cited as potential vectors of RVFV, even more than *Anopheles* spp. (Fontenille et al. 1998).

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access



No	Specimens	Type	Ct
1	177	*Aedes* /Site E	33.09
2	300	*Culex* /Site D	38.96
3	Positive Control	Positive Control	31.72
4	300	*Culex*/Site B	
5	300	*Culex*/Site A	
6	300	*Culex*/Site C	
7	8	*Aedes*/Site A	
8	Negative control	Negative control	
9	224	*Culex*/Site E	

Fig. 1. Real-time PCR for the detection of RVF virus inside pools of female mosquitoes.

RVFV was detected for the first time in Sudan in *Ae. vexans* with a higher level of viremia than in *Cx. quinquefasciatus* (cycle 30 versus cycle 34). These results coincide with studies conducted in Saudi Arabia, Kenya and Madagascar (Barry et al. 2002). Turell (2007) demonstrated that *Cx. quinquefasciatus* could artificially maintain and transmit the virus. The high level of viremia in *Ae. vexans* indicates that the virus is transovarially transmitted in these insects and that the virus is likely established in susceptible animal hosts in Soba West in Sudan.

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

Detection of RVFV in *Culex* spp. in Hellat Kuku indicates that the virus is amplified in livestock in that area; thus, posing a risk of transmission of the virus to humans, as noted by Martin et al. (2008). Furthermore, *Cx. quinquefasciatus* has been suggested to have a role in human-to-human transmission of the virus (Moutailler et al. 2007).

The 3 fundamental components of the epidemiological cycle of RVF, namely the presence of virus, susceptible hosts and potential vectors, occur in Khartoum. The expected scenario of an outbreak of RVF in Khartoum is that heavy rainfalls in the region will trigger emergence of diapausing *Ae. vexans* mosquitoes in Soba West. Large numbers of adult mosquitoes will survive for more than 30 d, during which time most will feed solely on mammalian blood, whereas a few will feed on avian hosts and a few will feed on a mixture of hosts (Molaei and Andreadis 2006). The virus will be vectored among livestock hosts. Thus, the virus will be subsequently amplified in livestock and likely acquired by other mosquitoes including *Aedes, Culex* and *Anopheles*. These secondary vectors, as well as *Aedes* mosquitoes will consequently transmit the virus to humans. In Hellat KuKu, heavy rainfalls will trigger emergence of *Culex* mosquitoes. The high density of vectors accompanied with the high viremia will enhance the transmission of the virus from livestock to humans directly. Heavy rains can influence outbreaks of RVF annually or at least at intervals of 2 - 3 yrs (Martin et al. 2008); thus, an outbreak of RVF is expected to occur in Khartoum in fall of 2010 or at least 2011, depending on rainfall amounts and patterns and the time of the previous outbreak which occurred in 2007 (FMOH 2008, WHO 2007b).

Acknowledgments

We acknowledge Dr. El Fatih M. Malik for funding this research and reading an earlier version of this manuscript. Thanks also to Dr. Fathi M. A. El Rabaa and Mr. Osama M. El Maki for their advice, guidance and support during the study. Special thanks to Dr. Mubarak El Karsani and Miss Rehab El Ageb for facilitating and conducting the molecular tests. Thanks also to the Federal Ministry of Health (Sudan) and the University of Khartoum, Faculty of Science for approval of this research.

References Cited

Barry, R. M., M. S. Godsey, M. B. Crabtree, H. M. Savage, Y. Al-Mazrao, M. H. Al-Jeffri, A. M. Abdoon, S. M. Al-Seghayer, A. M. Al-Shahrani and T. G. Ksiazek. 2002. Isolation and genetic characterization of Rift Valley Fever Virus from *Aedes Vexans* arabiensis. Kingdom of Saudi Arabia. Emerg. Infect. Dis. 8: 1492-1494.

Beier, J. C. 1996. Frequent blood-feeding and restrictive sugar-feeding behavior enhance the malaria vector potential of *Anopheles gambiae* S.I. and *An. funestus* (Diptera: Culicidae) in western Kenya. J. Med. Entomol. 33: 613-688.

Clements, A. N. and G. D. Paterson. 1981. The analysis of mortality and survival rates in wild populations of mosquitoes. J. Appl. Ecol. 18: 373-399.

Favier, C., K. Chalvet-Monfray, P. Sabatier, R. Lancelot, D. Fontenille and M. A. Dubois. 2006. Rift Valley Fever in West Africa: the role of space in endimicity. Trop. Med. Int. Health 11: 1878-1888.

FMOH. 2008. Final report for the emergency committee of Rift Valley Fever. Federal Ministry of Health, Sudan, Oct 2007- Jan 2008, pp. 1-69.

Fontenille, D., M. Traore-Lamizana, J. Diallo, J. Thonnon, P. Digoutte and H. G. Zeller. 1998. New vectors of Rift Valley Fever in West Africa. Emerg. Infect. Dis. 4: 289-293.

Gad, A. M., M. H. Mosaad, E. Sharif, I. M. Mahmoud and L. W. Owen. 1987. Rift Valley Fever virus transmission by different Egyptian mosquito species. Trans. R. Soc. Trop. Med. Hyg. 81: 694-698.

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access

Gubler, D. J. 1998. Resurgent vector-borne diseases as a global health problem. Emerg. Infect. Dis. 4: 442-449.

Gubler, D. J. 2002. The global emergence/resurgence of arboviral diseases as public health problems. Arch. Med. Res. 33: 330-342.

Hopkins, G. H. 1952. Mosquitoes of the Ethiopian region: 1. larval bionomics of mosquitoes and taxonomy of Culicine larvae. Oxford Univ. Press, London.

Jupp, P. G. 2005. Mosquitoes as vectors of human diseases in South Africa. South Africa Fam. Pract 47(9): 68-72.

Jupp, P. G., A. A. Grobbelaar, P. A. Leman, A. Kemp, R. F. Dunton, T. R. Burkot, T. G. Ksiazek and R. Swanepoel. 2000. Experimental detection of Rift Valley Fever Virus by reverse transcription-polymerase chain reaction assay in large samples of mosquitoes. J. Med. Entomol. 37: 467-471.

Martin, V., V. Chevalier, P. Ceccato, A. Anyamba, L. de Simone, J. Lubroth, S. de la Rocque and J. Domenech. 2008. The impact of climate change on the epidemiology and control of Rift Valley fever. Rev. Sci. Tech. Off. Int. Epiz. 27: 413-426.

Molaei, G. and T. G. Andreadis. 2006. Identification of avian- and mammalian-derived blood meals in *Aedes vexans* and *Culiseta melanura* (Diptera: Culicidae) and its implication for West Nile Virus transmission in Connecticut, U.S.A. J. Med. Entomol. 43: 1088-1093.

Moutailler, S., B. Michèle and A. Failloux. 2007. Short report: efficient oral infection of *Culex pipiens quinquefasciatus* by Rift Valley Fever Virus using a cotton stick support. Am. J. Trop. Med. Hyg. 76: 827-829.

Nayar, J. K. and E. Van Handel. 1971. The fuel for sustained mosquito flight. J. Insect Physiol. 17: 471-481.

Nayar, J. K. and D. M. Sauerman Jr. 1975a. The effects of nutrition on survival and fecundity in Florida mosquitoes. part 1. Utilization of sugar for survival. J. Med. Entomol. 12: 92-98.

Nayar, J. K. and D. M. Sauerman Jr. 1975b. The effects of nutrition on survival and fecundity in Florida mosquitoes. Part 3. Utilization of blood and sugar for fecundity. J. Med. Entomol. 12: 220-225.

Scott, T. W. and A. C. Morrison. 2003. *Aedes aegypti* density and the risk of dengue-virus transmission, Pp. 25-32. *In* W. Takken and T.W. Scott (eds.), Ecological Aspects for Application of Genetically Modified Mosquitoes. Kluwer Academic Press.

Turell, M. J., J. S. Lee, J. H. Richardson, R. C. Sang, E. N. Kioko, M. O. Agawo, J. Pecor and M. L. O'Quinn. 2007. Vector competence of Kenyan *Culex zombaensis* and *Culex quinquefasciatus* mosquitoes for Rift Valley Fever Virus. J. Am. Mosq. Control Assoc. 23: 378-382.

Weidong, G. and J. Robert. 2004. Detection of probability of arbovirus infection in mosquito populations. Am. J. Trop. Med. Hyg. 71: 636-638.

World Health Organization 2007a. Rift Valley Fever in Sudan – update. Epidemic and Pandemic Alert and Response (EPR). http://www.who.int/csr/don/2007_12_21/en/index.html.

2007b. Fact sheet of Rift Valley Fever. WHO media centre. http://www.who.int/mediacentre/factsheets/fs207/en/index.html.

2008. EHA Sudan weekly highlights. Series of weekly report WHO/SUD. www.emro.who.int/sudan/pdf/eha_highlights_52_08.pdf.

Wilson, M. L. 1994. Rift valley fever virus ecology and epidemiology of disease emergence. Ann. N. Y. Acad. Sci. 740: 160-180.

Downloaded from https://prime-pdf-watermark.prime-prod.pubfactory.com/ at 2025-07-04 via free access