

## Surveillance study on Rift Valley Fever in Jazan region, Saudi Arabia

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### Abstract:

A cross-sectional survey was performed during the rainy season from July to December 2013, to investigate the circulation of RVF virus serologically among animal species within RVF outbreak zones of 2000. The sero-surveillance including, a total of 4930 serum samples collected from local herds, sentinel animals, and smuggled animals(1372,1670,1888) respectively. These sera were assayed for immunoglobulin M (IgM), and immunoglobulin G (IgG) antibodies against RVF disease by using a commercially enzyme-linked immunosorbent assay(ELISA). Of 1375 sera sampled from local herds, RVF was diagnosed in two clinically healthy animals. Despite, no serological evidence of RVF was found in smuggled animals, two out of 1670 investigated sentinel animals, were found positive for RVF IgG antibodies. The estimated infection rate in local herds was 0.15%, while (0.12%) was in sentinel herds. Interestingly, the detection of Thirty-nine IgG antibodies within zones where vaccination against RVF is not practiced and at the same time close to others where vaccination is performed, clearly demonstrated the need for marker vaccines that enable differentiation between vaccinated and naturally infected animals(DIVA). In conclusion, there was a recent circulation of RVF virus in Almekhwa and Alarda district. These finding recognized both districts as high risk zones. Consequently, special consideration should be given to those regarding control measures.

## Introduction

Rift valley fever (RVF) is a serious infectious disease of both domestic ruminants and humans, caused by mosquito-borne virus belongs to the family Bunyaviridae, genus phlebovirus [1]. Virus transmission usually occurs either through the bite of an infected mosquito or by direct contact with infected animal tissues, body fluids or aborted fetal materials [2]. Despite the virus has a potential to infect a wide range of animals including goats, cattle, camels, dogs, cats, and ferrets, it has been recognized as acute and fatal disease in new born lambs and causes high rates of abortion in pregnant ewes as well [3]. Since the disease was first isolated in Kenya 1931, several outbreaks affected animals and humans have been reported in much of sub-saharan Africa, mainly and more frequent in Kenya, Somalia and Tanzania [4]. By the year (2000) a significant turning point for the disease has occurred, the virus emerged in new geographical areas outside of African continent, extended to Arabian Peninsula affected Yemen and Saudi Arabia, where a total of (886) human cases were reported in Saudi Arabia [5]. This outbreak raised concerns about the potential incidence and establishment of the disease in different environmental conditions and could affect areas haven't ever experienced the disease before [6].

During this outbreak, Jazan region-South west Saudi Arabia- has been the hardest hit by the disease. (65.6%) of animal cases occurred in Jazan,(26.9%) in Asir and (7.5%) in Alquenfeda. The infection rate was (23%, 8.7% and 2%) in Jazan, Asir and Alquenfeda respectively [7]. Apparently, the virus seems to be introduced to Saudi Arabia during the religious festivals(Eid Aladha) through importation of live animals from African Horn countries, as long as, the virus isolated from the first patients during 2000 outbreak had an RNA sequence similar to the virus isolated in 1997-1998 East African outbreaks [8],[9]. Additionally, it has been Proved that,

Saudi Arabia was free from RVF until 1995 and most probably before the 2000 epidemic [10].

A comprehensive control program has been implemented to minimize the spread of the disease, Since the disease was first recognized in the region. The control strategies involving livestock vaccination with live attenuated vaccine, restriction of animal movement, insecticide land spray and aerial spray, as well as, active surveillance and virus detection in mosquitoes by molecular techniques. These control measures seem to be effective and still have played a considerable role in disease control, since no clinical disease has been reported yet in humans and animals[11].

Recently, (RVF) disease has been persistent in Jazan region and sporadic cases continue to occur [12]. Furthermore, a favorable habitat for the disease has always been constituted in Jazan region during the rainy season. Consequently, surveillance programs should be continued to prevent the recurrence of outbreaks and monitoring trends. In this paper, we carried out a sero-surveillance study during the 2013 rainy season, involving, clinical and serological examination for local herds, sentinel animals, and smuggled animals which had been kept at Altwal-quarantine station( the border between Saudi Arabia and Yemin), to investigate the potential existence of RVF virus circulation in livestock within disease surveillance zone that had been identified during the 2000 outbreak. The results of this study will be useful in identifying high risk zones, as well as enhancing both of control program and surveillance system.

## **Materials and Methods**

### **Area of Study and Population**

Jazan region is located in South-west Saudi Arabia between longitudes 42 43 East and latitudes 16 17 North, near the Yemeni borders which represent the southern and eastern border. The Red sea borders the region from the west and extended

(300) Km along the sea coast and Asir region from the north. The region covers an area of 40.457 KM<sup>2</sup>, the terrain of the region is varied consisting of mountain, costal and fertile plains [13]. The existence of both ecological diversity pattern and different types of vegetation, listed the region as one of the richest areas in Saudi Arabia with animal biodiversity[14]. It is noteworthy that, several seasonal valleys extended from mountains constitute alluvial flood plains as well as a considerable amounts of rainfall in the region( 800mm/Y). Furthermore, changes have been made in Wadi system by orienting the water flow to irrigate fields through channel systems creating many large and small water pools[15]. However, these conditions resulting in good breeding habitats for RVF vectors.

### **Study Design and Data Collection**

A cross-sectional survey was carried out during rainy season from August 2013 to December 2013, to investigate RVF virus circulation in animal's blood within previously affected zones. The study area involving 18 districts belong to Jazan region, and Tohamat Aseer with routine vaccination program, as well as four districts belong to Tohamat Albaha and Mecca hadn't experienced vaccination against RVF disease. The serological survey including serum samples from local herds, sentinel animals and smuggled animals at Altwal quarantine. A total of 1375 blood samples were taken within a range of 10-15% from every herd with one of the following clinical signs (abortion-diarrhea-fever-death in young animals). While 1670 sera from sentinel herds were collected monthly.

Sentinel animals are about 275 animals at the same age and related to the local breeds. These animals were imported in 2004 from regions free from RFV infection and subjected to IgM and IgG antibodies tests against RVF disease before placed on eleven districts that previously affected by 2000 outbreak: Alarda-Alhurath–Abuareesh- sabia- Almasarha- Bulgazi –Baish- Mahaeel- Mejardah-Mekhwa- quenfeda. Twenty-five animals were located on each site and have not

been vaccinated yet. While Smuggled animals those were introduced illegally to the region from Yemeni borders through border villages and seized by security officers and transferred to Altwal quarantine where they subjected to quarantine majors.

### **Samples Collection:**

Whole blood samples (8.0 mL) collected into SST tubes (BD) from sheep and goats with one of the following clinical signs (abortion-diarrhea-fever-death in young animals). Vaccinated animals within 45 days before the study were not included. Sera from sentinel herds were collected monthly during the study period. The blood allowed to clot at room temperature for 30 minutes, centrifuged at 3000g for 15 minutes, and the serum aliquoted into new vials, labeled and stored at -80 C.

### **Laboratory Examinations:**

#### **Serological Tests:**

IgG-sandwich and IgM-capture enzyme-linked immunosorbent assay (ELISA) used for the detection and quantitation of (IgG) and (IgM) antibodies to RVF in animals sera [16]. The commercial ELISA kits were performed according to the producer's description.

#### **Data Analysis:**

The serological data were analyzed with SPSS software (Statistical Package for the Social Sciences) version 20, IBM/SPSS. The following variables for each sample were recorded: sex, species, place, IgM, and IgG. Subsequently, cross-tabulation and the X chi square test used to examine the relationships between variables.

### **Results**

In this study a total of 4930 serum samples were collected from local herds, sentinel animals and smuggled animals at Altwal quarantine: 1372, 1670, 1888

respectively. These samples were tested for IgM and IgG antibodies against RVF virus. Serum samples of local herds were collected from different administrative districts including: Leeth 203, Almekhwa 281, Golwa 599, Ardiat 152, Baish 99, Sabia 31, and Mhaeil 7. Most of these samples 821 (59.8%) were goats, 543(39.6%) were sheep, while only eight (0.58%) camels. Out of 1372 sera, only two samples tested positive for IgM antibodies against RVF disease without clinical signs, in two herds at AL-Mekhwa district. The infection rate in local herds was 0.15%. Additionally, a total of 76 serum samples tested positive by Sandwich ELISA for IgG antibodies. The IgG antibodies ranged from 0.1% in Leeth to 2.5% in Baish(Table I).

**Table I**  
**Seroprevalence of Rift Valley Fever in local herds**

Place	IgM		IgG		Species			Total
	negative	positive	negative	positive	sheep	goat	camel	
Leeth	203	0	201	2(0.1%)	129	66	8(0.58%)	203
Almekhwa	279	2(0.1%)	273	8(0.6%)	100	181	0	281
Golwa	599	0	597	2(0.1%)	208	391	0	599
Ardiat	152	0	125	27(2.0%)	40	112	0	152
Baish	99	0	65	34(2.5%)	63	36	0	99
Sabia	31	0	28	3(0.2%)	2	29	0	31
Mhaeil	7	0	7	0	1	6	0	7
Total	1370	2(0.1%)	1296	76	543	821	8(0.58%)	1372

**Table II**

**The results of testing sentinel herds sera**

Place	IgG		Species		Total
	negative	positive	sheep	goat	
<b>Alarda</b>	113	2(0.1%)	53	62	115(6.9%)
<b>Abuareesh</b>	159	0	145	14	159(9.5%)
<b>Sabia</b>	147	0	141	6	147(8.8%)
<b>Almasarha</b>	203	0	131	72	203(12.2%)
<b>Bulgazu</b>	172	0	109	63	172(10.3%)
<b>Alhurath</b>	145	0	113	32	145(8.7%)
<b>Baish</b>	209	0	141	68	209(8.9%)
<b>Mahaeel</b>	125	0	100	25	125(7.5%)
<b>Quenfeda</b>	149	0	100	49	149(8.9%)
<b>Mekhwa</b>	100	0	76	24	100(6.0%)
<b>Mejardah</b>	146	0	82	64	146(8.7%)
<b>Total</b>	1668	2(0.1%)	1191	479	1670

**Sentinel Animals**

A total of 1670 serum sample from sentinel animals were assayed for (IgM), and (IgG) antibodies against RVF. Among these animals only two samples belong to Alarda herd had reacting IgG antibodies to RVF disease without clinical signs. The infection rate was 0.12% (Table II).

### **Smuggled Animals:**

Screening of smuggled animals revealed that none of the samples (1888), collected from Altwal quarantine during the study period were found positive for IgM antibodies against RVF virus.

### **Discussion, Finding, and Recommendations**

Routine vaccination for susceptible herds at the age of six month has still been practiced extensively in Jazan region and Aseer region, as well as Alquenfedat district, while four districts (Leeth - Almekhwa - Glowat-Ardiat ) had no vaccination program and considered as monitoring area confined to vector control and surveillance. Among those four districts two cases in two herds tested positive for IgM antibodies against RVF disease. The infection rate was estimated at (0.15%). This is lower than previously reported in 2004 surveillance (0.36% ) [17] and much lower comparing to that reported during 2000 outbreak (23%) [18].

A herd visit conducted to investigate each infected herd in Almekhwa, revealed that one herd with 70 animals and the other one consisted of 180 animals sheep and goats. Both two herds didn't show any signs of abortion, no deaths among lambs and all animals were clinically healthy as well as no new animals have recently entered the herd. However, there were still some epidemiological evidences justified and supported claims for the existence of recent virus circulation in Almekhwa district, most notably, Nawan dam located nearby at distance of 500 Mitre from the infected herds, which provides an ideal habitat for RVF vectors. Additionally, as no vaccination against RVF is practiced in these districts, it was suggested that the IgM antibodies were due to natural infection. Furthermore, RVF disease was diagnosed in Almekhwa sentinel herd five months prior to study period. Anyhow, these cases of RVF disease without clinical signs were similar to those reported in Sudan during 2007 outbreak, when the disease was not demonstrated in livestock even after the human cases did[19].

In sentinel herds, on the other hand, two positive cases (IgG) were reported in Alarda herd without clinical signs too. It is worth mentioning that, the same herd tested positive for IgM antibodies while routine inspection for sentinel herds, which had been done five months before the current study. The recurrence of infection in Alarda district is most likely attributed to several ecological circumstances favoring the development of vector populations and contribute to virus cycling in domestic animals: firstly, it is noteworthy that, the district was heavily affected by the disease during 2000 outbreak; and secondly the area occupied by *Aedes* mosquitoes who responsible for the continued presence of RVF virus in enzootic foci, as the female mosquito able to transmit the virus to her offspring through eggs leading to new generation of infected mosquitoes[20]. Finally, the district has experienced heavy rainfall over the last three years(200 ML), these heavy rainfall flooded the breeding sites of *Aedes* spp resulting in, very large numbers of mosquitoes[21], [22]. However, these results are in contrast with that reported by [23]. who reported that the region was free from RVF infection for that time.

As no routine vaccination has been practiced in : Alardiat, Golwa and Leath since 2005, the demonstration of 39 IgG antibodies raises a number of questions about whether these antibodies were due to natural infection or vaccinated animals imported from other districts where annual vaccination is practiced. It could not be definitely confirmed, since both of two possibilities supported by evidences : these antibodies could be due to natural infection as long as the areas free from vaccination since 2005 and it has been proved that IgG antibodies following vaccination against RVF disease disappeared after the elapse of six years and eleven months [24]. On the other hand, short distances less than 100 Km separated between those districts and vaccinated areas, supported the possibility of interference between two areas, notably the current sandwich ELIZA cannot

distinguish vaccinated from naturally infected animals. This situation clearly point the need for new generation of vaccines enable the distinction between vaccinated and naturally infected animals [25].

It was concluded that RVFV circulates sub-clinically in different ecological systems in Alarda districts (high grassland with high rainfall) and Almekwa district(dry grassland). Study finding recognized the two areas as critical zones pose serious threats to animal and human population and emphasize the need for enhancing vector control activity and raising herd immunity in Alarda district to prevent the recurrence of infections in those districts.

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