Detection of Crimean Congo Haemorrhagic Fever virus markers in ticks in Upper Guinea-Republic of Guinea.

ABSTRACT

The aim of this study was to map the distribution of agents carrying Crimean Congo haemorrhagic fever virus (arbovirus-tica) in the natural region of Upper Guinea. Two types of analysis methods (RT-PCR and ELISA) were used. Of a total of 578 ticks collected and divided into 254 pools, the genus Amblyomma was the most frequently encountered with 83 tick pools. Molecular analysis (RT-PCR) revealed 2 positive cases (0.8%). Enzyme-linked immunosorbent assays yielded only one positive case (0.4%). We found that the species Rhipicephalus geigyi was the main vector and reservoir of the pathogen in Upper Guinea.

Key words: CHF Congo virus, tick pools, RT-PCR, ELISA, Upper Guinea.

INTRODUCTION

CCHF was first identified in 1944, during an epidemic of haemorrhagic fevers among Russian servicemen in the Crimea. The aetiology of the epidemic was quickly suspected to be arboviral. This hypothesis was confirmed after the injection of filtrates of ticks of the genus Hyalomma marginatum in 1965 reproduced the same symptoms in volunteers, and the virus was thus named "Crimean virus" (Bente et al., 2013; Hoogstraal, 1979; Whitehouse, 2004). In 1969, it was discovered that the Crimean virus was antigenically identical to a virus isolated in the Democratic Republic of Congo in 1956, known as the Congo virus, which caused disease similar to that caused by the Crimean virus. The virus was therefore renamed "Crimean-Congo haemorrhagic fever virus" (Bente et al., 2013; Hoogstraal, 1979; Whitehouse, 2004).

Crimean Congo haemorrhagic fever (CCHF) is an acute viral infection (arthropozoonosis) transmitted by ticks of the genera Hyalomma, Amblyomma and Rhipicephalus (reservoir and vector), endemic throughout Africa, Asia and the Middle East (L. Gétaz, L. Loutan, N. Mezger ; 2012 ; D Baumann, N Pusterla O Péter ; 2003 ; K. Senior; 2008). Infection occurs through the bite of infected ticks or direct contact with the blood and tissues of infected humans or animals. Hospital staff, slaughterhouse workers, livestock farmers and people exposed to viremic animals and their ticks are all groups at ri The World Health Organization (WHO) has announced that a new case of Crimean-Congo haemorrhagic fever (CCHF) has occurred in Mauritania in Kithat in the Wilaya of Guidimakha. A total of seven contacts (four family members and three health workers) have been identified (WHO Africa Office; 2019).

METHODOLOGY

The Upper Guinea region was used as the study area. This prospective and descriptive study was carried out at the Institut de Recherche enBiologieAppliquée de Guinée (IRBAG) from June to December 2022. The biomaterial consisted of tick pools. Enzyme-linked

immunosorbent assay (ELISA) and molecular RT-PCR were used to search for markers of this virus.sk.

MAP OF GUINEA

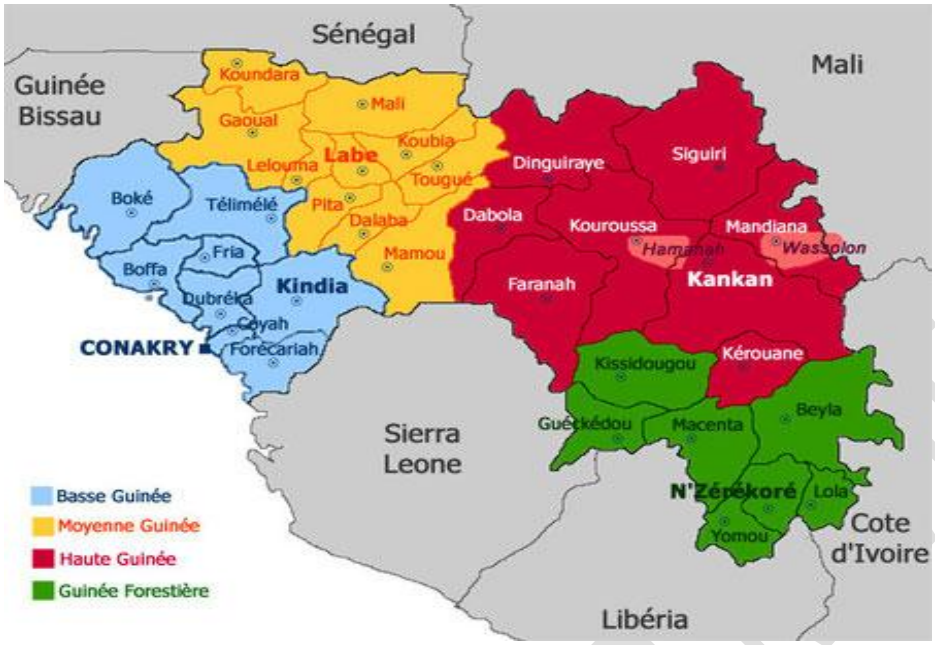

Fig 1. Study area

RESULTS

Table 1: Number of positive cases in Upper Guinea by prefecture and according to the two analysis methods.

Prefectures	Pool name	Positive cases			
		RT-PCR	(%)	ELISA	(%)
Faranah	90	1	0.4	0	0
Dabola	58	0	0	0	0
Kankan	106	1	0.4	1	0.4
Total	254	2	0.8	1	0.4

In Upper Guinea, tick collection yielded a sample of 254 pools distributed as follows: 106 in Kankan, 90 in Faranah and 58 in the prefecture of Dabola; analysis of the results revealed 2 positive cases. One case was found in Kankan and the other in Faranah, with a rate of 0.8%. No positive cases were found in Dabola. As regards the ELISA enzyme-linked immune sorbent assay, we found only 1 positive case in the prefecture of Kankan, which proves the sensitivity of the molecular method compared with the ELISA enzyme-linked immune sorbent assay.

Table 2: Positivity rate of the different tick species collected in the Faranah prefecture using the RT-PCR method.

Tickspecies	Pool name	Positive cases		
		PCR	(%)	$IC_{95\%}$
Am varigeatum	29	0	0	-
Hy trucatum	5	0	0	-
Rh annulatus	7	0	0	-
Rh decoloratus	33	0	0	-
Rh geigyi	15	1	0.4	0.5-0.3
Rh senegalensis	1	0	0	-
Total	90	1	0.4	0.5-0.3

This table showing the distribution of tick species in the Faranah prefecture shows that out of a total of 90 tick pools collected, only one positive case was found using the RT-PCR analysis method, i.e. 0.4%. No cases were found using the enzyme-linked immune sorbent assay. The species concerned was Rhicipicephalis geigyi.

Table 3: Positivity rate of the different tick species collected in the prefecture of Kankan by the analysis methods.

Tickspecies	**Pool name**	**Positive cases**			
		PCR	**(%)**	**ELISA**	**(%)**
Am varigeatum	35	0	0	0	0
Hy trucatum	8	0	0	0	0
Rh annulatus	7	0	0	0	0
Rh decoloratus	36	0	0	0	0
Rh geigyi	18	1	0.9	1	0.9
Rh senegalensis	2	0	0	0	0
Total	106	1	0.9	1	0.9

Observation of the results of the two analysis methods (RT-PCR and ELISA) shows a similarity in terms of the species concerned (Rhicipicephalis geigyi) and the positivity rate (0.9%).

Table 4: Positivity rate of the different tick species collected in the prefecture of Dabola by the analysis methods.

Tickspecies	Pool name	Positive cases		
		ELISA	(%)	IC
Am varigeatum	19	0	0	-
Hy trucatum	5	0	0	-
Rh annulatus	6	0	0	-
Rh decoloratus	20	0	0	-
Rh. geigyi	8	0	0	-
Rh. senegalensis	0	0	0	-
Total	58	0	0	-

From this table, we can see that of the 58 tick pools collected in the prefecture of Dabola, no positive case was detected by the two RT-PCR and ELISA analysis methods, but it should be noted that all the species encountered in the other two prefectures are represented in Dabola.

DISCUSSION

Molecular analysis (RT-PCR) revealed 2 positive cases (0.8%). Enzyme-linked immunosorbent assays yielded only one positive case (0.4%). We found that the species Rhipicephalus geigyi was the main vector and reservoir of the pathogen in Upper Guinea.

A study carried out in Guinea by a team of Russo-Guinean researchers led by E.V Naidenova, in 2020, on the prevalence of Crimean-Congo haemorrhagic fever virus in rural areas of Guinea, showed that among the ticks studied, the estimated prevalence of CCHFV was 1.3 ± 0.4%. Five of the eight tick species studied were identified as carriers of CCHFV in Guinea (E.V Naidenova, al., 2020).

These results are superior to our own (0.4% by the enzyme-linked immunosorbent assay and 2 positive cases (0.8%) by the molecular method (RT-PCR).

The results found by F. Farhadpour et al. in 2016 in southern Iran show a detection rate of 4.5% for 9 samples examined, and the species concerned were Hyaloma marginatum, Hyalomaanatolicum and Rhicicephalussanguinus; these results are higher than those found in Upper Guinea by both analytical methods. In our case, we had not encountered the genus Hyaloma as a carrier of the CHFCongo virus.

The results obtained by a team of researchers (Aillen E. et al., 2016) working on the sero-surveillance of pathogenic viruses circulating in West Africa indicate a higher result than ours, using the molecular RT-PCR method and enzyme immunoassay, with a frequency of the Crimean Congo haemorrhagic fever virus of 2%.

Our results can be compared with studies carried out by Sanidad M. in 2017 in Spain, which show a geographical breakdown similar to that of Guinea; the results obtained mention 3.2% as the prevalence obtained. This result is higher than that found in our study.

CONCLUSION

As a result of the research carried out and the results obtained, CCHF virus antigen was detected in 1 tick pool and virus RNA in 2 tick pools. All the tick pools in which viral antigen was detected contained CCHF virus RNA. Positive results were obtained in two prefectures (Kankan and Faranah). We found that the species Rhipicephalus geigyi was the main vector and reservoir of the pathogen in Upper Guinea.

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