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Epidemiology of Crimean-Congo hemorrhagic fever in Senegal: temporal and spatial patterns

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Summary. Aspects of the spatial and temporal patterns of transmission of Crimean-Congo hemorrhagic fever (CCHF) virus were studied in Senegal, West Africa. A country-wide serological survey of domestic animals indicated that transmission was most intense in the northern dry sahelian zone and least in the southern, more humid guinean zone. Human IgG prevalence, ranging from nearly 20% to <1% among 8 sites throughout the region, also was greatest in the north. A fatal human case of CCHF from Rosso, Mauritania in 1988 was studied and an accompanying serosurvey of human contacts and domestic animals indicated epidemic transmission during that period. Systematic samples of adult ixodid ticks on domestic animals allowed us to analyze the distribution and relative abundance of potential CCHF virus vectors, demonstrating that *Hyalomma* spp. predominated in those biotopes where transmission was most intense. A prospective study of CCHF virus infection and tick infestation in sheep exposed a period of epizootic transmission in 1988 that corresponded temporally with increased abundance of adult *H. truncatum* and *H. impeltatum.* Four strains of CCHF virus were isolated from pools of these ticks and of *Rhipicephalus guilhoni.* Our results suggest that CCHF virus is focally endemic throughout the region, although highly variable in time and space, and that the relative abundance of *Hyalomma* ticks may be the primary determinant of epidemic transmission.

Introduction

The distribution of Crimean-Congo hemorrhagic fever (CCHF), although irregular and focal, is remarkably widespread. CCHF virus circulates throughout much of the southern Soviet Union, central Asia, southern Europe, the Middle East, and Africa [45], covering three of the world's seven major biotic zones [18]. Zoonotic disease in humans has been recognized in about a dozen countries, but serological or virological evidence of CCHF virus transmission has been found throughout these regions. At least 30 species of ticks [3] and more than 20 different vertebrate species [45] have been shown to be infected. Despite the abundance of research documenting the widespread distribution, diversity of possible vectors and numerous potential vertebrate reservoirs for CCHF virus, our understanding of the transmission cycle(s) remains incomplete. We can only speculate, at present, as to the importance of variables that maintain transmission and stimulate epizootics or epidemics.

In sub-saharan West Africa, CCHF virus and antibodies have been detected in ticks, domestic and wild vertebrates, or humans from Benin [10], Burkina Faso [10, 37], Mauritania [34–36], Nigeria [4, 8, 44] and Senegal [9, 29–33]. Initial observations in Senegal during the late 1960's demonstrated indirect evidence of domestic animal and tick infections [5]. During the 1970's and early 1980's, researchers at the Pasteur Institute in Dakar isolated numerous strains of CCHF virus from ticks feeding on cattle and sheep at Senegal slaughterhouses [29–33]. Antibodies were detected at sites throughout the country and a human case and antibodies in animals were documented along the border in Mauritania [36]. Thus, CCHF virus has been circulating in Senegal during at least the past 2 decades.

The factors that influence transmission of CCHF virus include the density of competent vector ticks, particularly of the genus *Hyalomma*, and the relative abundance of vertebrates that serve as both hosts to these ticks and as possible reservoirs of CCHF virus [18]. Sustained transmission is found only where *Hyalomma* ticks are present and epizootic or epidemic transmission is believed to occur during periods of increased abundance of these ticks [45]. However, the general relationship between tick species diversity and the magnitude of CCHF virus transmission has not been systematically studied within a particular geographic region. While the vertebrate host associations of these ticks typically are well-defined [18], the role that most of these vertebrates play in the natural maintenance cycle or amplification of CCHF virus remains enigmatic.

Human cases of CCHF have been recognized from more than a dozen countries on 3 continents [45]. In sub-saharan Africa at least 69 human cases of CCHF have been documented, mostly in southern Africa. In West Africa, however, only two non-fatal human cases have been previously reported, this despite a relatively high seroprevalence indicating a high prevalence of

human infections. The first case, in 1983, was from Selibaby, Mauritania near the northeastern border with Senegal [36]. Another case later that year was reported from southeastern Burkina Faso [37]. Thus, the true incidence of human infection and particularly of human disease remains unknown. We report here on various studies designed to investigate the spatial and temporal aspects of CCHF virus transmission within this region of West Africa. Specifically, we present summaries of studies on the seroprevalence of sheep and of humans [47], a recent fatal case [11], the distribution of potential vectors, an epizootic corresponding with changes in tick abundance, and recent virus isolations from ticks.

Study sites and methods

A prospective, multidisciplinary research program based at the Pasteur Institute in Dakar, Senegal was begun in 1987 to investigate various aspects of the ecology and epidemiology of CCHF virus transmission in Senegal, Gambia, and Mauritania. This region, encompassing a variety of habitats and ecological zones, ranges from the Sahara desert in the north to humid forests in the south. The biota changes primarily in association with differences in average annual rainfall ranging from 200 mm in the northern sahelian zone to 1500 mm in the southern sudano-guinean and sub-guinean zones [23]. The diversity and abundance of potential vector ticks is also large [2, 3, 12], as is the vertebrate fauna of potential hosts [21].

Serological studies

In order to define the distribution and intensity of CCHF virus transmission, and the environmental factors associated with such infection in Senegal, a systematic serosurvey of IgG antibodies among sheep was undertaken [47]. These abundant and widespread domestic ungulates are infested by potential tick vectors and express antibodies to CCHF virus infection. Furthermore, because their average age is only a few years, sheep seroprevalence serves as an index of relatively recent transmission. Blood samples were obtained between November 1987 and February 1988 from herds chosen in 26 randomly selected administrative districts throughout Senegal [47]. Age was estimated by dental examination and grouped into four categories: <14, 14–28, 29–36, and >36 months. Sera were frozen in the field and later tested at the Pasteur Institute in Dakar for IgG antibody to CCHF virus.

Human serological studies were based on IgG prevalence from samples collected during 1986–88 from 8 different sites in the region. In addition to our samples from Yonofere, Dahra, Bandia, and Kedougou, we tested sera collected as part of other studies from Dagana and Ziguinchor by Dr. Alain Jouan, from Tambacounda by Dr. Jean-Francois Saluzzo, and from Rosso by Dr. Elizabeth Manus. The details of the sampling methods varied among the sites; however, all sera were obtained from apparently healthy people who were asked to donate blood for medical research. People were bled by venipuncture and sera were frozen at $-20\,^{\circ}C$ from 1 to 4 days later and tested as above.

Sera were tested for evidence of anti-CCHF virus IgG using an ELISA test [28] modified slightly by adding a saturating solution of PBS with 0.05% Tween 20 and 1% non-fat bovine milk. In this direct ELISA test, 96-well plates (Immulon II, Dynatech Laboratories, Alexandria, VA) were coated with diluted CCHF virus hyperimmune mouse ascitic fluid. CCHF virus (strains IbAr 10200 from Sokoto, Nigeria and the recently isolated Dak H49199 from a human in Rosso, Mauritania) in crude suckling mouse brain was heat inactivated at

60°C for 1 h. and then added. Test sera, diluted 1:400, followed by test-species specific anti-immunoglobulin conjugated with horse radish peroxidase (Biosys, Compiegne, France) was used to detect the IgG. A chromogenic substrate (orthotoluidine, Sigma, LaVerpilliere, France) was added for colorimetry. All plates included a control of crude suckling mouse brain without CCHF virus antigen. Differences in optical density (OD) between the test and control wells were measured at 450 nm using an automatic reader (Multiscan MCC/340, Flow Laboratories, Irvine, Scotland) coupled to a microcomputer. By iterations of the distribution of OD values, we determined the mean of the population of negatives. Sera were considered positive if the OD was greater than 3 standard deviations above the mean of negatives.

IgM antibodies were detected by immunocapture ELISA [38]. Plates were coated with anti-μ chain specific for the species being tested. The test serum, followed by CCHF viral antigen was then added. The detecting antibody was a high-titered mouse ascitic fluid against CCHF virus antigen. Anti-mouse immunoglobulin conjugated with horse radish peroxidase and the chromogenic substrate were added as above. Evaluation and criteria were as for IgG.

Virus and antigen identification

Virus isolation was attempted by intracranial inoculation of suckling mice and by inoculation of Vero cells using undiluted and 10-fold diluted sera. Virus identification was made by an indirect immunofluorescent test on Vero cells, using polyclonal and monoclonal antibodies. The identity of virus isolates was confirmed by a complement fixation test at the WHO collaborating Center for Arboviruses at the Pasteur Institute in Dakar.

An antigen capture ELISA [38] was also employed to test for presence of CCHF virus antigen in human sera. Plates first were coated with anti-human IgM μ chain specific antibody, followed by human sera with high titer IgM. The test serum was added next and then a high-titered anti-CCHF virus monoclonal IgG was added to bind to any antigen captured from the test serum. An anti-mouse IgG, conjugated with horse radish peroxidase and the chromogenic substrate were used for colorimetry.

Vector tick distribution and abundance

The relative abundance of potential vector ticks was studied by analyzing monthly samples of adult ticks attached to domestic animals in various sites throughout Senegal. Sheep, goats or cattle were deticked monthly throughout at least 1 year in order to avoid bias due to seasonal variation in tick activity. We sampled 3 sites in northern Senegal: Yonofere, Dahra, and Bandia (Fig. 1). In Yonofere, 5 herds of sheep were chosen monthly by chance encounter from May 1987 through May 1989 and 10 randomly selected individuals from each herd were sampled. All ticks were removed with forceps and stored for later identification and virus isolation. The same regime was followed in Dahra from May 1987 through August 1989. In Bandia, 12 sentinel goats and 2 cattle were examined similarly 3 times each month from April 1987 through March 1989.

In addition, published data from other sites in Senegal were also analyzed. Sites were compared by calculating the mean number of ticks per host (cattle, sheep, or goats) from monthly samples of 40 hosts made during 12 or more months during 1985–1989.

Temporal relation between tick abundance and virus transmission

To determine the relationship between tick abundance and CCHF virus transmission, sheep were bled periodically and simultaneously examined for the presence of ticks. Approxi-

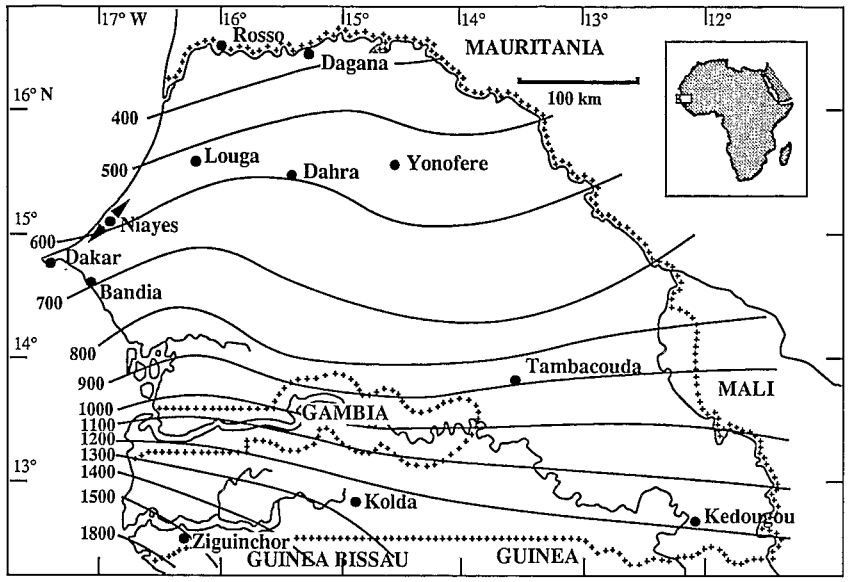

Fig. 1. Map of Senegal showing sites where observations were made, and isoyeths indicating the average annual rainfall [23]. Surrounding countries are noted

mately 300 individually-identified sheep were bled every 2 or 3 months during 1987–1989 at the Centre de Recherches Zootechniques at Dahra, as were another 200 sheep from free-ranging herds in the village of Yonofere (Fig. 1). We estimated the prevalence of infection and attempted to correlate this with variation in tick abundance and other environmental variables.

Efforts to isolate virus were undertaken using ticks collected monthly from domestic ungulates in Yonofere and Bandia during 1987 through 1989. In addition to ticks from above mentioned collections, herdspeople from these and surrounding villages were given tubes for collecting ticks that they removed from their animals. Ticks were frozen at −70 °C until being pooled by species and herd, ground in Hanks' solution, centrifuged, and inoculated into suckling mice or cell culture.

Results

Spatial distribution of animal seroprevalence

A total of 942 sheep were bled between November 1987 and February 1988 from the 22 administrative regions that were studied [47]. The overall prevalence of IgG antibodies among sheep was 10.4%. Antibody prevalence was equal for male and female sheep, but increased with age from 2.1% of the youngest animals to 18.2% among the oldest age group [47].

The spatial pattern of CCHF virus transmission, as indicated by antibody prevalence, varied among the villages. Prevalence appeared highest in the north, decreasing to nil in the south. When seroprevalence was organized into 5 ecologic zones [27], three-quarters of the sheep tested from the northernmost, Sahelian zone showed evidence of previous infection, a rate

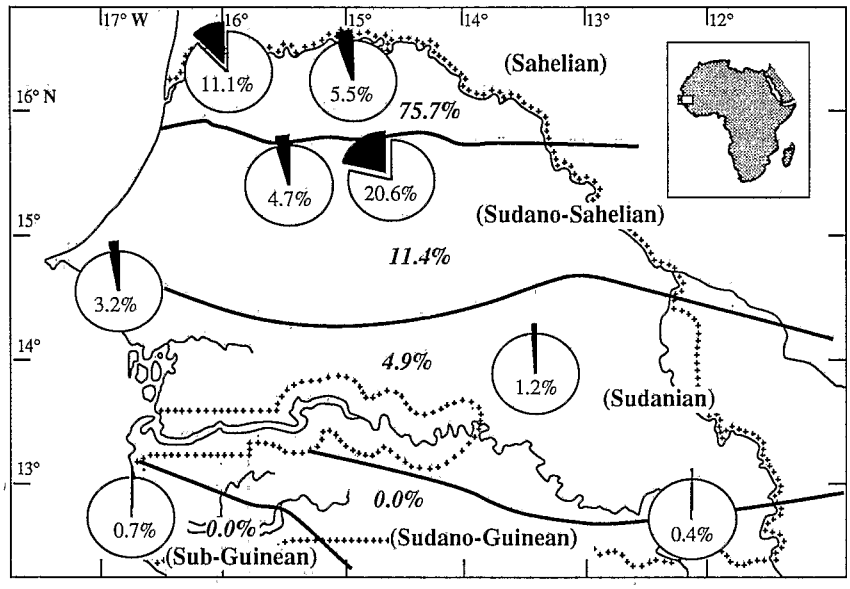

Fig. 2. Prevalence of IgG antibodies to CCHF virus in sheep and humans throughout Senegal. Human IgG prevalence is indicated by the circles that correspond to villages where samples were made. IgG prevalence in sheep is shown in italics as the average for all sheep sampled in each of 5 bioclimatic zones that are named in parentheses

which decreased to zero toward the southern sudano-guinean and sub-guinean zones (Fig. 2). Furthermore, antibody prevalence correlated negatively with rainfall, declining from a maximum of 31.3% at the 400 mm isohyet to 0% at the 1200 mm isohyet ($r^2 = 0.74$, $n = 9$, $p < 0.003$). Those regions experiencing the least rainfall had the highest rate of CCHF virus transmission. Attempts to explain this spatial pattern using geological formations, soil types, altitude, and sub-surface water revealed no such relationships. Thus, CCHF virus transmission was found to occur with greatest intensity in dryer, more sparsely vegetated Sahelian and northern Sudanian regions.

Comparison with results from previous serosurveys revealed that antibody prevalence for the sheep in our study varied within the range of average values reported from other sites in Africa (Table 1). The negative correlation between antibody prevalence and rainfall observed in our study was consistent with results from the 1969 Senegal survey by Chunikhin et al. [5] (Table 1). CCHF virus transmission in Senegal, particularly in the north, appears to have been especially intense during the 1980's.

Human seroprevalence

The prevalence of IgG for people aged 10 or more varied by as much as an order of magnitude among the villages sampled (Fig. 2). In northern sites along the Senegal River, 17 of 150 (11.3%) people from Rosso, and 5 of 91

Table 1. Prevalence of anti-CCHF virus antibodies among domestic ungulates from various sites in Africa

Country	Site	Domestic animal	Prevalence seropositive	No. tested	Ref.
Egypt	Wadi Natroun	C	14.3%	21	[7]
	Qena	C	13.3	13	[7]
	Cairo	S	18.2	66	[7]
	Cairo	D	8.8	34	[7]
Kenya	Eldoret	C	7.3	55	[1]
	Isiolo	C	8.3	96	[1]
	Kajiado	C	1.1	92	[1]
	W. Pokot	C	3.0	100	[1]
Mauritania	Selibaby	C	32.0	25	[35]
Nigeria	Bauchi	C	26.6	64	[44]
	Niger	C	29.5	535	[44]
	Kaduna	C	22.0	565	[44]
Senegal	(north)	C	11.5	235	[5]
	(central)	C	6.2	113	[5]
	(south)	C	15.4	26	[5]
	(s. west)	C	8.6	93	[5]
	(north)	S	5.8	512	[5]
	(central)	S	1.4	70	[5]
	(s. west)	S	0	70	[5]
	(central)	G	0	80	[5]
	(s. west)	G	1.4	70	[5]
Uganda	Mbarara	C	36.5	104	[19]
R.S.A.	Bloemhof	C	64.1	170	[43]
	(east)	C	26.5	6128	[40]
	(north)	C	28.4	8667	[41]
	Bloemhof	S	27.4	270	[43]
Zimbabwe	(north)	C	45.0	763	[41]

Domestic animals are cattle (*C*), sheep (*S*), dromedaries (*D*), and goats (*G*)

(5.5%) people from Dagana were seropositive. In the north-central Sahel plains, 5 of 106 (4.7%) and 27 of 128 (21.1%) residents from Dahra and Yonofere, respectively, had IgG antibody. In Bandia, further south along the coast, 3 of 92 (3.2%) adults tested showed evidence of previous infection. Low IgG prevalence was found in southern sites including 1 of 140 (0.7%) in Ziguinchor, 1 of 84 (1.2%) in Tambacounda, and 2 of 225 (0.9%) in Kedougou. The prevalence of human antibody against CCHF virus was highest in the north and lowest in the south, a pattern that corresponded to that of sheep.

Human disease

Sera from patients presenting with symptoms of hemorrhagic fever during May 1988 at the hospital in Rosso, Mauritania, on the northern Senegal border, were tested for antibody to CCHF virus and for antigen [11]. Among 8 such patients, 6 showed positive IgG titers; 3 of these 6 patients died. In one fatal case, IgM antibodies were detected and a strain of CCHF virus was isolated.

Subsequent investigations of people in 3 nomadic camps from which patients came showed that of 99 people tested, 36 (36%) had IgG antibodies, including 1 with IgM [11]. Among 17 sheep and goats that were tested in 2 villages, 5 (29%) had IgG and 1 (6%) also had IgM antibody. In addition, 46% of 120 such animals were infested with adult *Hyalomma impeltatum* or *H. dromedarii* ticks (data not shown). Thus, a period of intense transmission of CCHF virus, including human infection and disease occurred in the region around Rosso.

To determine whether such transmission occurred in other parts of the region, a survey of sheep from various sites in southern Mauritania was undertaken; IgG antibody rates ranged between 10% and nearly 50% [11]. Furthermore, about 7% of these sheep had IgM antibodies; intense transmission of CCHF virus apparently occurred throughout much of southern Mauritania during March through July 1988.

Vector tick distribution and abundance

The predominant ixodid ticks in our 3 northern Senegal study sites were *Hyalomma truncatum*, *H. impeltatum* and *Rhipicephalus guilhoni* (Table 2). These three species represent more than 95% of ticks infesting domestic animals in Dahra and Yonofere, and more than 80% of such ticks in Bandia. *H. marginatum rufipes*, *Amblyomma variegatum*, and other *Rhipicephalus* spp. comprised the remaining collections.

We reanalyzed studies by Gueye and collaborators [13–17], to compare their results with ours and again found that *Hyalomma* ticks were most abundant in sites of northern Senegal (Table 2). *Rhipicephalus evertsi evertsi*, and other *Rhipicephalus* spp. occurred predominantly in central and southern sites, such as Tambacounda and Kolda, while *Amblyomma variegatum* and *Boophilus* spp., absent in the north, were most abundant in the south. The distribution and relative abundance of ticks from our studies were similar to that described by Morel [25] in 1958 (Table 3).

Temporal relation between tick abundance and virus transmission

The prevalence of IgM in sheep at Dahra remained at less than 10% from September 1987 through February 1988, rising to nearly 40% in May 1988

Table 2. Infestations of adult ticks on domestic ungulates from various sites in Senegal

Zone region	Host[a]	Sample period	No. ticks		Percentage of[b]													Ref[c]
			total	mean	Ht	Hr	Hi	Hd	Hs	Re	Rs	Rg	Ru	Rl	Av	Bd	Bg	
Sahelian																		
Yonofere	S	5/87–5/89	4195	4.4	50.9	0.9	0.3	—	—	—	—	45.1	—	—	—	—	—	
Dahra	S	5/87–5/89	4218	4.1	44.1	0.1	51.2	<0.1	—	0.3	—	4.3	—	—	—	—	—	
Louga	S	1/84–12/84	1817	3.0	—	—	1.4	—	—	98.6	—	—	—	—	—	—	—	[13]
	G	1/84–12/84	97	0.2	—	—	4.1	—	—	95.9	—	—	—	—	—	—	—	[13]
Sudano-sahelian																		
Bandia	C	4/87–3/89	1439	10.0	55.4	16.2	—	—	1.7	0.3	—	25.6	—	—	0.7	0.1	—	
	G	4/87–3/89	743	1.0	11.7	—	—	—	—	0.3	—	87.6	—	—	0.1	0.3	—	
Niayes	C	4/82–9/83	25315	35.2	30.0	8.2	—	—	1.3	<0.1	3.8	—	<0.1	—	27.0	29.1	—	[17]
	G	4/82–9/83	909	1.3	13.5	—	—	—	—	<0.1	<0.1	3.5	17.4	—	57.8	6.7	—	[17]
Sudanian																		
Tambacounda	C	10/83–12/84	2173	3.6	16.0	74.5	—	—	—	<0.1	<0.1	—	<0.1	6.1	2.8	<0.1	—	[14]
	S	10/83–12/84	126	0.2	10.3	2.4	—	—	—	84.9	—	—	—	2.4	—	—	—	[14]
	G	10/83–12/84	32	0.1	15.6	—	—	—	—	65.6	—	—	—	18.8	—	—	—	[14]
Sudano-guinean																		
Kolda	C	1/86–3/87	9647	16.1	9.9	1.1	—	—	—	<0.1	<0.1	—	3.9	15.1	34.1	—	35.8	[15]
	S	1/86–3/87	207	0.3	4.4	0.1	—	—	—	—	—	—	10.1	10.6	25.6	—	9.2	[15]
	G	1/86–3/87	42	0.1	19.0	—	—	—	—	7.1	—	—	7.1	23.8	2.4	—	40.5	[15]

[a] Hosts are cattle (*C*), sheep (*S*), and goats (*G*)

[b] Tick species are *Hyalomma truncatum* (Ht), *H. marginatum rufipes* (Hr), *H. impeltatum* (Hi), *H. dromedarii* (Hd), *H. impressum* (Hs); *Rhipicephalus evertsi evertsi* (Re), *R. senegalensis* (Rs), *R. guilhoni* (Rg), *R. sulcatus* (Ru), *R. lunulatus* (Rl); *Amblyomma variegatum* (Av); *Boophilus decoloratus* (Bd), *B. geigyi* (Bg)

[c] Previously published work is cited in References. Our data from Yonofere, Dahra and Bandia are original

—Species not collected

Table 3. Relative abundance of ticks feeding on domestic ungulates in various bioclimatic zones in Senegal

Climatic zone[a]	Average annual precip.	Ticks[b]												
		Hd	Hi	Hr	Hs	Ht	Av	Bd	Bg	Re	Rg	Rs	Rm	Ru
Sahelian	250–500 mm	++	+++	++	+	+					o			
		×	××	×		×××	×			×	×××			
N. Sudan.	500–1000		+	+++	+	+++	++	++		+	++		+	
			×	××	×	×××	××	×		×	×××		×	
S. Sudan.	1000–1250			+	o	++	+++	+	++[c]			++o	++	++
				×		×	×××		××	×				
Guinean	>1250						+++	o	++			+++		+++

Relative abundances include: very abundant (+++), moderately abundant (++), rare (+), extremely rare (o), absent (blank)

\+ Values from Morel [25]

× Values from our research [46; unpubl.]

[a] Zones and precipitation from Morel [25]

[b] Tick species include *Hyalomma dromedarii* (Hd), *H. impeltatum* (Hi), *H. marginatum rufipes* (Hr), *H. impressum* (Hs), *H. truncatum* (Ht); *Amblyomma variegatum* (Av); *Boophilus decoloratus* (Bd), *B. geigyi* (Bg); *Rhipicephalus evertsi evertsi* (Re), *R. guilhoni* (Rg), *R. senegalensis* (Rs), *R. muhsamae* (Rm), *R. sulcatus* (Ru)

[c] P. C. Morel, pers. comm.

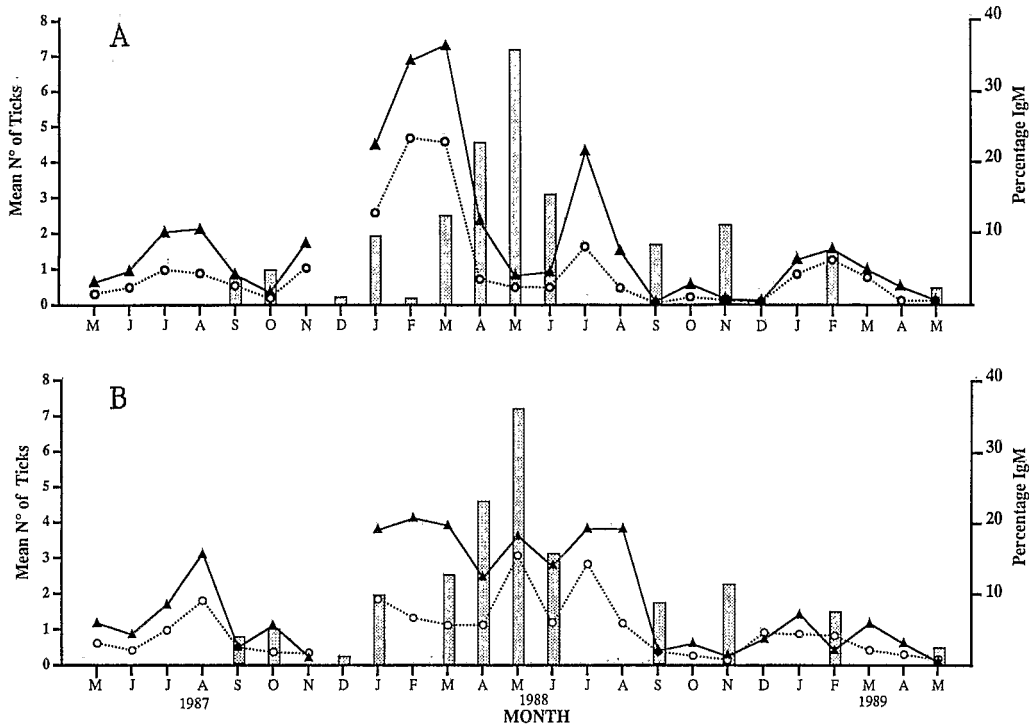

Fig. 3. Average number of adult male (▲—▲) and female (○ · · · ○) *H. truncatum* (**A**) and *H. impeltatum* (**B**) on sheep sampled in Dahra, Senegal from May 1987 to May 1989, and the monthly prevalence of IgM antibodies to CCHF virus in sheep

(Fig. 3). IgG prevalence varied from 5% to 21%, rising in correspondence with that of IgM to a high of more than 60% in September 1988, then declining. In all, roughly two-thirds of more than 300 sheep were infected during this epizootic. Concurrently, numerous sheep became sick and a few died. Two of 4 sheep bled because they were ill had high titer IgM antibodies.

Adult tick abundance on sheep, simultaneously monitored throughout this same period, also changed (Fig. 3). A rapid increase in the abundance of adult *Hyalomma impeltatum* and *H. truncatum* feeding on sheep was observed prior to and during the period of increased transmission. Abundance of these ticks declined at about the same time that IgM prevalence diminished. The other tick that heavily infested sheep in this region, *Rhipicephalus guilhoni*, was absent at that time (data not shown). The prevalence of IgM among sheep in Yonofere remained essentially unchanged at less than 5% during the entire period.

Virus isolation from vector ticks

From January 1988 through June 1989, a total of 31,630 ticks of eight species were grouped into 1,838 pools and tested for arboviruses (Table 4). In addition to Wad Medani (57 strains), Dugbe (2), and Bandia (1) viruses, four

Table 4. CCHF virus isolations during 1988 and January through June, 1989 in ticks removed from cattle, sheep, and goats in Yonofere, Dahra, and Bandia, Senegal

Site	No. (lots) of ticks[a] tested							
	Ht	Hr	Hi	Hs	Re	Rg	Av	Bd
Yonofere								
1988	9755 (501)	342 (38)	131 (17)	—	1 (1)	4373 (231)[b]	—	—
1989	4095 (236)	128 (23)	5 (4)	—	3 (3)	981 (76)	—	—
Dahra								
1988	3260 (174)	27 (9)	3972 (212)[b]	—	31 (9)	104 (9)	—	—
1989	564 (37)	17 (3)	343 (30)	—	5 (5)	48 (9)	—	—
Bandia								
1988	327 (41)	130 (26)	—	10 (6)	5 (4)	653 (50)	80 (14)	2 (2)
1989	106 (19)[c]	53 (19)	—	1 (1)	—	97 (29)	—	—
Total	19766 (1008)	1019 (118)	4451 (263)	11 (7)	45 (22)	6256 (404)	80 (14)	2 (2)

[a] Tick species are *Hyalomma truncatum* (Ht), *H. marginatum rufipes* (Hr), *H. impeltatum* (Hi), *H. impressum* (Hs); *Rhipicephalus evertsi evertsi* (Re), *R. guilhoni* (Rg); *Amblyomma variegatum* (Av); *Boophilus decoloratus* (Bd)

[b] One strain of CCHF virus isolated

[c] Two strains of CCHF virus isolated

strains of CCHF virus were isolated. The CCHF virus strains include one from a pool of 17 male *H. impeltatum* taken from sheep at Dahra in March 1988, one from 20 male *Rhipicephalus guilhoni* removed from sheep in Yonofere during August 1988, and two strains from male–female pools of 9 and 10 *H. truncatum* removed from cattle at Bandia.

Discussion

The prevalence of antibodies in sheep varied among our study sites from intense to nil, suggesting that CCHF virus transmission was either spatially focal or temporally sporadic. In some sites, the prevalence of IgG indicated that nearly half of all sheep were infected at least once during their lifetime. Using an estimate of 32 months as the average age of sheep [47] 1 in 10 or 20 sheep become infected each year in sites where prevalence was high. This is likely an underestimate, as transmission probably was not temporally uniform. Intense epizootics of CCHF virus seem likely.

Transmission of CCHF virus was most intense in the northern, arid Sahel region of Senegal and decreased consistently toward the more moist, southern forest zones. A similar biogeographic relationship has been observed elsewhere in regions of enzootic transmission; sites in Africa, Eurasia, and the Middle East that experience the most intense transmission tend to be

relatively more arid [45]. Transmission in Africa occurs primarily in arid savannah grasslands characterized by long dry seasons, and where *Hyalomma* ticks abound. In Eurasia, regions dominated by deserts, semi-deserts and steppes similarly support circulation of this virus. Perhaps the distribution and abundance of potential vectors, which are influenced by climatic conditions, in particular rainfall [2, 3], in turn determine the distribution of CCHF virus. Similarly, biogeographic zones differ in the presence and abundance of potential vertebrate reservoir(s) which may be capable of maintaining horizontal transmission. Alternatively, the periodic, long-distance migration of domestic animals that occurs in drier regions of Africa might increase exposure to questing ticks, thereby elevating the prevalence of infection. Whether greater transmission in semi-arid zones is due to differences in the species diversity of vector ticks, suitable reservoirs, amplifying vertebrate hosts, or other variables deserves further study.

Antibody prevalence increased with the age of sheep in those regions where transmission was relatively intense suggesting that exposure there was enzootic. However, our cross-sectional sample was not large enough to document local epizootics retrospectively. That antibodies were found in sheep of all ages in many regions suggests endemic transmission throughout much of the country. The absence of antibodies in our sample of sheep from southern Senegal probably reflects periodic or focalized transmission at a level too low for us to detect.

Human seroprevalence varied spatially in a manner similar to that of sheep. However, people are exposed, on average, during more years than are sheep, and people are more likely to be sampled other than where they had been exposed. Despite these differences, the pattern of human IgG prevalence also was greatest in the north and least in the south. Human risk seems to correspond with that of domestic animals [45], which may serve as useful sentinels of human infections.

The isolation of CCHF virus from a patient with a hemorrhagic fever and the detection of low titer IgM suggested that this person died of Crimean-Congo hemorrhagic fever, the first reported fatal case from West Africa [11]. Two other patients died with similar symptoms but showed no detectable IgM and low IgG titers. Thus, for the latter 2 patients, we did not determine that their fatal clinical syndromes were due to CCHF virus infection. Among contacts of the hospitalized cases, one family member had IgM antibody and more than one-third of contacts showed evidence of past CCHF virus infection. By comparison, a 1984 sample of healthy people from the same area indicated an IgG prevalence of 5.5% [35]. Furthermore, domestic animals throughout the region exhibited IgM consistent with recent transmission. The combination of recognized human cases, IgM and high prevalence IgG among case contacts, and IgM among domestic animals suggest that recent transmission of CCHF virus occurred in the region during May 1988, producing human infection and disease.

The paucity of human cases of CCHF in Senegal and Mauritania is enigmatic in light of seroprevalence rates indicating transmission at levels equal to or greater than that found in other regions of the world [45]. In southern Africa, at least 23 primary human cases (10 fatal) have been documented since 1981 [41], while seroprevalence among humans there was only 1.5% [40]. Many cases have been diagnosed in the Soviet Union, yet antibody prevalence is similarly low [45]. The hypothesis that West African strains of CCHF virus are less pathogenic to humans than strains in Eurasia or southern Africa has been suggested, but differences in the availability of health care and surveillance make such a hypothesis difficult to test. Access to clinical and diagnostic services is scarce in north-central Senegal thus limiting the correct identification of people with severe hemorrhagic fevers. Deaths in remote villages occur unreported or may be reported without etiology. These factors would lead to underestimating the amount of severe disease and number of deaths due to CCHF, though to what extent we cannot determine. Because CCHF virus appears weakly- or non-pathogenic for ungulates and disease in humans is rarely diagnosed, only large-scale epidemiological studies will accurately determine the true intensity of virus circulation and human disease.

The vector(s) of CCHF virus in Senegal remain poorly defined, although our results indicate that one or more *Hyalomma* spp. are important. Other studies have suggested a correlation between *Hyalomma* tick abundance and virus transmission [18], despite the fact that ticks from 6 other genera have been shown to be infected [3]. Our results demonstrated a positive correlation between the spatial patterns of *H. truncatum* and *H. impeltatum* abundance, and of the prevalence of infection in humans and sheep. Although *Amblyomma variegatum* and *Rhipicephalus* spp. were present where evidence of transmission was found, these ticks were more abundant in the central and southern sites where CCHF virus circulation was less. The predominance of *Hyalomma* spp. where transmission was most intense in humans and in sheep suggests that these ticks may serve both as enzootic and endemic vectors. That CCHF virus previously has been isolated from 2 other *Hyalomma* spp. as well as from ticks from 3 other genera [3] suggests that other ticks may also play a role in transmission.

The distribution pattern and relative abundance of ticks in Senegal, described originally in 1958 by Morel [25], was similar to that which we determined from our studies [46] (Table 3). *Hyalomma* spp. predominate in the dry Sahelian zone becoming progressively less abundant in the semi-arid Sudanian zone and the more humid habitats of the guinean zone. *Amblyomma variegatum* and *Boophilus* spp. predominate in the Sudanian and Guinean zones. Curiously, *Rhipicephalus guilhoni*, confused with *R. sanguineus* before 1962 [26], has apparently increased in relative abundance in the Sahelian zone since the 1950's. In general, the geographic distribution and relative abundances of these ticks exhibited a pattern that could be classified by bioclimatic zone.

A temporal relationship between the abundance of *H. truncatum* and *H. impeltatum* and increased transmission further supports our observation that these ticks play a role as important vectors. Tick abundance and anti-CCHF IgM prevalence in sheep were both relatively low, then rose precipitously, and later declined during a 2-year period of study. This focal epizootic in northern Senegal could not have resulted from transmission by other ixodid ticks, as only *Rhipicephalus guilhoni* occurs in abundance there and its distinct seasonal pattern peaked after most transmission had occurred [3]. In addition, virus transmission was apparently intense in other areas of southern Mauritania during this same period, as suggested by the high IgM prevalence in our sheep serosurvey [11]. Curiously, no epizootic was observed 150 km east in Yonofere despite the fact that adult *H. truncatum* but not *H. impeltatum* abundance, simultaneously rose to a high level there. Whether tick abundance, tick infection rates or other factors influenced this difference is not known.

H. truncatum appears to be effective as a vector of CCHF virus [24, 39] and certain *Hyalomma* ticks may also serve as reservoirs, in that transovarial transmission in *H. marginatum rufipes* and *H. marginatum marginatum* has been experimentally demonstrated [20, 22, 48]. Amplifying, horizontal transmission may be limited to a short period of viremia during which infected and uninfected ticks are feeding simultaneously. It seems unlikely that adults of these ticks horizontally transmit to immatures because they feed on different hosts. In the absence of transovarial transmission, domestic animals may play no role in the CCHF virus cycle, other than as a food resource for tick reproduction. Nevertheless, migration by these domestic animals, small mammal or bird population fluctuations, or other bioclimatic changes could lead to the observed temporal and spatial heterogeneity of CCHF virus transmission.

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