Crimean–Congo hemorrhagic fever (CCHF) is a tickborne zoonotic disease that is characterized by hemorrhagic fever and can progress from mild, nonspecific signs to a severe and fatal hemorrhagic disease. The CCHF virus (CCHFV) is an enveloped, segmented, negative-sense, single-stranded RNA member of the family *Nairoviridae*, genus *Orthonairovirus*. CCHFV has been detected in >35 species of ticks worldwide, among which ticks belonging to the genus *Hyalomma* are the primary vectors in humans and wild and domestic animals (1). Humans are infected through tick bites and direct contact with infected blood and body fluids during occupational exposure (e.g., farming, slaughtering, and medical and nursing care).

CCHF is endemic in Africa, Asia, and the Balkan region (2). In Western Europe, autochthonous human cases were reported only in Spain, where CCHFV was identified in *H. lusitanicum* ticks (3). In Corsica, a French Mediterranean island, 9.1% of livestock (i.e., cattle, goats, sheep) serum samples contained CCHFV-specific IgG during 2014–2016 (4). Entomologic surveys revealed that the *H. marginatum* tick, a vector of CCHFV, was present in Corsica (5).

The Study

To assess whether CCHFV circulates in Corsica, we collected 8,051 ticks from wild and domestic animals in selected sites on the island during 2016–2020 (Table, Figure). These 8,051 ticks included 7,156 ticks taken from 3,674 domestic animals and 895 ticks taken from 188 wild animals. They consisted of 4,177 *Rhipicephalus bursa* (51.8%), 2,386 *H. marginatum* (29.6%), 839 *Dermacentor marginatus* (10.4%) and 282 *H. scupense* (3.5%) ticks. We identified ticks at the species level by using a pictorial guide and confirmed morphologic identification by using sequencing of mitochondrial 16S rDNA (5). We then pooled up to 10 ticks per pool on the basis of developmental stage (nymphs, non-engorged females, and male adults) and host (Table). Pools, containing an average of 2.5 ticks (range 1–10 ticks) were crushed in phosphate-buffered saline with TissueLyser II (QIAGEN, https://www.qiagen.com) at 5,500 rpm for 3 min. We spiked each pool before extraction with a predefined amount of MS2 bacteriophage to monitor the subsequent steps (nucleic acid extraction, reverse transcription, and PCR amplification) and to detect the presence of inhibitors and enzymatic reactions as described (6). We performed DNA extraction by using QIAcube HT and a QIAamp cador Pathogen Mini Kit (QIAGEN), according to the manufacturer’s instructions. We eluted DNA in 150 μL of buffer and stored at −20°C. We tested each pool for the presence of CCHFV RNA by using a real-time, reverse transcription PCR (7) and

Author affiliations: Laboratoire de Virologie, Université de Corse–Institut National de Santé et de la Recherche Médicale, Corte, France (V. Cicculli, A. Maitre, A. Falchi); Unité des Virus Emergents, Aix Marseille Université, Marseille, France (V. Cicculli, N. Ayhan, X.N. de Lamballerie, R. Charrel); Laboratoire de Recherches sur le Développement de l’Élevage, Institut National de la Recherche pour l’Agriculture, l’Alimentation et l’Environnement, Corte (J.-C. Paoli); Campus International de Baillarguet, Montpellier (L. Vial); Parc Naturel Régionale de Corse, Corte (S. Mondoloni)

DOI: https://doi.org/10.3201/eid2805.211996
the presence of Nairovirus RNA by using a pangene-
ic reverse transcription PCR (8).

We detected neither CCHFV RNA nor Nairo-

virus RNA in the 8,051 ticks. The absence of CCHFV

or Nairovirus RNA was not attributable to tech-
nical problems or presence of inhibitors, which were

ruled out by MS2 bacteriophage monitoring. More-

over, we detected viral RNA corresponding to new
tickborne Phleboviruses in 40 samples (5%) and Flavi-
virus in 7 samples (0.9%); these samples remain un-
der investigation, and results will be reported after
detailed characterization.

Conclusions

We considered whether CCHFV RNA was not de-
tected because of a low minimum infection rate (MIR)
that a larger number of ticks would have been re-
quired. We calculated the theoretical power that could
be achieved by using the number of ticks obtained in
our study. On the basis of an expected CCHFV preva-

lence \( P \approx 0.2\% \) and a pool size \( k \) of 2 ticks, a total

of 7,676 ticks have to be tested for a prevalence esti-
mation with a 95% CI and a precision \( d \) set at \( \pm 0.001 

because the disease prevalence is \(<0.1 (10\%) (9). \) Thus,

with a sample of 8,051 ticks, we were able to detect a

prevalence of \( \geq 0.2\% \).

The rate of CCHFV-infected ticks in countries

in Europe with enzootic foci ranges from 0.50% to

3.70% among Hyalomma spp. ticks (2.8% [44/1,579

H. lusitanicum ticks] in Spain, 3.7% [6/161 H. margin-
atum ticks] in Bulgaria, and 0.5% [1/199 H. margin-
atum ticks] in Kosovo) and from 1.5% to 6.2% among

Rhipicephalus spp. (1.5% [2/123 R. sanguineus ticks] in

Bulgaria and 6.2% (8/130 R. bursa ticks) in Koso-

vo) (10). Other studies conducted outside of Europe

have largely reported MIR values >0.2% among
ticks: 0.71% in South Africa (1.6% [15/914] H. trunca-
tum and 0.2% [2/1,149] H. rufipes) (11); 2.6% in Mau-
ritania (39/1,517 Hyalomma spp.) (12); 3.8% (20/525

Hyalomma spp.) in Pakistan (13); and 51.5% (103/200

H. marginatum) in Turkey (14). These studies were

conducted during the past 5 years using methods

comparable to those of our study. The number of

Hyalomma (n = 2,682) and Rhipicephalus (n = 4,177)
ticks that we tested are much higher than reported in

these previous studies. Therefore, our study would

Table. Ticks collected, by host, number of ticks, and number of tick pools, in a study of Crimean–Congo hemorrhagic fever virus in ticks from wild and domestic animals, Corsica, France, 2016–2020

| Host and tick species | No. ticks | No. tick pools |
|-----------------------|-----------|---------------|
| Cattle, n = 1,211     |           |               |
| *Rhipicephalus bursa* | 3,413     | 818           |
| *Hyalomma marginatum* | 1,343     | 475           |
| *H. scupense*         | 282       | 96            |
| *Boophilus annulatus* | 130       | 47            |
| *Ixodes ricinus*      | 85        | 33            |
| *H. punctata*         | 14        | 10            |
| *R. sanguineus*       | 96        | 32            |
| *Dermacentor marginatus* | 2   | 2             |
| Total                 | 5,365     | 1,513         |
| Horses, n = 201       |           |               |
| *H. marginatum*       | 1,026     | 247           |
| *R. bursa*            | 637       | 135           |
| *R. sanguineus*       | 27        | 10            |
| Total                 | 1,690     | 392           |
| Wild boar, n = 182    |           |               |
| *D. marginatus*       | 837       | 222           |
| *H. marginatum*       | 13        | 7             |
| *R. bursa*            | 9         | 6             |
| *I. ricinus*          | 13        | 5             |
| *R. sanguineus*       | 1         | 1             |
| Total                 | 873       | 241           |
| Sheep, n = 773        |           |               |
| *R. bursa*            | 101       | 93            |
| Total                 | 101       | 93            |
| Deer, n = 4           |           |               |
| *R. bursa*            | 9         | 4             |
| *H. marginatum*       | 4         | 1             |
| Total                 | 13        | 5             |
| Mouflon sheep, n = 2  |           |               |
| *R. bursa*            | 8         | 5             |
| *I. ricinus*          | 1         | 1             |
| Total                 | 9         | 6             |
| Overall               | 8,051     | 2,250         |
have been able to recognize CCHFV presence for a prevalence ≥0.2%, which is 10 times lower than the lowest overall prevalence value reported to date in countries where CCHF is present: 2.1% (95% CI 1.3%–2.9%) according to a recent meta-analysis (10). Furthermore, another study addressing the presence of CCHFV RNA in *Hyalomma* spp. ticks (362 *H. marginatum* and 135 *H. scupense*) and *Rhipicephalus* ticks (*n* = 518) collected in 2014 from domestic and wild animals in Corsica also provided only negative results (15). In all countries where CCHF cases are described, the observed MIR of ticks is ≥2.5 times higher than the detection limit in our study (0.2%). Another argument that strongly supports the contention that the lack of detection of CCHFV or *Nairovirus* RNA was not caused by technical problems is based on the consideration that the protocol used in this study enables the detection of a wide variety of different CCHFV strains, a fact that confirms the accuracy of the results (7,8).

Recent studies determine whether CCHFV is present in Corsica and to what extent it is a threat for human populations, provide contrasting data. On one hand, tick species that are able to transmit CCHFV are present and widely distributed, and a serologic study based on ELISA screening and neutralization test for confirmation supports the presence of CCHFV or an antigenically related agent. On the other hand, the absence of detection of CCHFV RNA (or an antigenically related agent) in a large number of ticks, together with the absence of a CCHF case, supports the absence of CCHFV in Corsica to date.

In any case, the absence of a documented case of CCHF together with the lack of detection of CCHFV RNA in tick species that are recognized as a competent vector enables us to declare that Corsica is not a hotspot for CCHFV and that the threat to the human population is very limited. However, this discrepant set of data pleads for a One Health approach for dealing with the CCHF question in Corsica and the potential exposure of island population. To do so, the roadmap established by the World Health Organization’s R&D blueprint (https://www.who.int/teams/blueprint/about) should be followed. Because the accuracy of CCHFV serologic assays has been questioned, several tests must be combined as advocated. Then, serologic studies in animals and humans must be synchronized with virus detection in ticks and systematic screening of patients with uncharacterized febrile illness during the tick season. A need exists for a large-scale One Health prospective program for surveillance of ticks, vertebrates, and humans in Corsica.

**Figure.** Locations of tick collection sites (for cattle and horses) for a study of Crimean–Congo Hemorrhagic fever virus in ticks from wild and domestic animals, Corsica, France, 2016–2020.

**Acknowledgments**
We are grateful to the staff of the slaughterhouse at Ponte-Leccia for their help in collecting ticks from cattle.

This study has been funded in part by the European Virus Archive Global Project, which has received funding from the European Union’s Horizon 2020-INFRAIA-2019 research and innovation programme (grant agreement no. 871029) and by the European Union–Cepheid Innovative Medicines Initiative Joint Undertaking (as part of the Viral Haemorrhagic Fever: Modern Approaches For Developing Bedside Rapid Diagnostics [VHFMoDRAD] under grant no. 823666).
About the Author

Mr. Cicculli is a doctoral student at the University of Corsica and Aix-Marseille University. His research interests include the epidemiology of vectorborne pathogens.

References

1. Spengler JR, Bergeron É, Rollin PE. Seroepidemiological studies of Crimean-Congo hemorrhagic fever virus in domestic and wild animals. PLoS Negl Trop Dis. 2016;10:e0004210. https://doi.org/10.1371/journal.pntd.0004210

2. Portillo A, Palomar AM, Santibáñez P, Oteo JA. Epidemiological aspects of Crimean-Congo hemorrhagic fever in Western Europe: what about the future? Microorganisms. 2021;9:649. https://doi.org/10.3390/microorganisms9030649

3. Negredo A, Sánchez-Ledesma M, Llorente F, Pérez-Olmeda M, Belhassen-García M, González-Calle D, et al. Retrospective identification of early autochthonous case of Crimean-Congo hemorrhagic fever, Spain, 2013. Emerg Infect Dis. 2021;27:1754–6. https://doi.org/10.3201/eid2706.204643

4. Grech-Angelini S, Lancelot R, Ferraris O, Peyrefitte CN, Vachiery C, Pedarrieu A, et al. Molecular Detection of Spotted-Fever Group Rickettsiae in ticks collected from domestic and wild animals in Corsica, France, 2014–2016. Emerg Infect Dis. 2020;26:1041–4. https://doi.org/10.3201/eid2605.191465

5. Cicculli V, Oscar M, Casabianca F, Villechenaud N, Charrel R, de Lamballerie X, et al.; Molecular Detection of Spotted-Fever Group Rickettsiae in ticks collected from domestic and wild animals in Corsica, France. Pathogens. 2019;8:138. https://doi.org/10.3390/pathogens8030138

6. Ninove L, Nougairede A, Gazin C, Thirion L, Delougou I, Zandotti C, et al. RNA and DNA bacteriophages as molecular diagnosis controls in clinical virology: a comprehensive study of more than 45,000 routine PCR tests. PLoS One. 2011;6:e16142. https://doi.org/10.1371/journal.pone.0016142

7. Wölfer R, Paweska JT, Petersen N, Grobbelaar AA, Leman PA, Hewson R, et al. Virus detection and monitoring of viral load in Crimean–Congo hemorrhagic fever virus patients. Emerg Infect Dis. 2007;13:1097–100. https://doi.org/10.3201/ eid1307.070068

8. Lambert AJ, Lanciotti RS. Consensus amplification and novel multiplex sequencing method for S segment species identification of 47 viruses of the Orthobunyavirus, Phlebovirus, and Nairovirus genera of the family Bunyaviridae. J Clin Microbiol. 2009;47:2398–404. https://doi.org/10.1128/JCM.00182-09

9. Ausvet. Epitools—epidemiological calculators. 2016 [cited 2022 Feb 19]. https://epitools.ausvet.com.au

10. Belobo J, Msimang V, Weyer J, le Roux C, Kemp A, Burt F, Tempia S, et al. Risk factors associated with exposure to Crimean-Congo haemorrhagic fever virus in animal workers and cattle, and molecular detection in ticks, South Africa. PLoS Negl Trop Dis. 2021;15:e0009384. https://doi.org/10.3201/eid2706.204643

11. Msimang V, Weyer J, le Roux C, Kemp A, Burt F, Tempia S, et al. Risk factors associated with exposure to Crimean-Congo haemorrhagic fever virus in animal workers and cattle, and molecular detection in ticks, South Africa. PLoS Negl Trop Dis. 2021;15:e0009384. https://doi.org/10.3201/eid2706.204643

12. Schulz A, Barry Y, Stock S, Pickin MJ, Ba A, Chitimia-Dobler L, et al. Detection of Crimean-Congo hemorrhagic fever virus in blood-fed Hyalomma ticks collected from Mauritanian livestock. Parasit Vectors. 2021;14:542. https://doi.org/10.1186/s13071-021-04819-x

13. Kasi KK, von Arnim F, Schulz A, Rehman A, Chudhary A, Onew M, et al. Crimean-Congo haemorrhagic fever virus in ticks collected from livestock in Balochistan, Pakistan. Transbound Emerg Dis. 2020;67:1543–52. https://doi.org/10.1111/tbed.13488

14. Akyildiz G, Bente D, Keles AG, Vatansever Z, Kar S. High prevalence and different genotypes of Crimean-Congo hemorrhagic fever virus genome in questing unfed adult Hyalomma marginatum in Thrace, Turkey. Ticks Tick Borne Dis. 2021;12:101622. https://doi.org/10.1111/tbed.13488

15. Grech-Angelini S, Stachurski F, Vaysier-Taussat M, Devillers E, Casabianca F, Lancelot R, et al. Tick-borne pathogens in ticks (Acari: Ixodidae) collected from various domestic and wild hosts in Corsica (France), a Mediterranean island environment. Transbound Emerg Dis. 2020;67:745–57. https://doi.org/10.1111/tbed.13593

Address for correspondence: Vincent Cicculli, Laboratoire de virologie, UR7310 Université de Corse, 20250, Corte, France; email: cicculli_v@univ-corse.fr