DRAFT EPI-2: TRIP REPORT*

ASHLEY L. GREINER

This is a preliminary report of an EPiAID investigation. Future correspondence, reports or publications related to this investigation may present results, interpretations, and recommendations that differ from those contained in this document.

ABSTRACT

From January to September 2014, the National Centers for Disease Control and Public Health (NCDC) identified 22 cases of Crimean-Congo hemorrhagic fever (CCHF), including three deaths, in the country of Georgia. CCHF is endemic in this region, but this is the highest annual number of cases reported since passive surveillance for this Category A bioterrorism agent was initiated in 2009. In this investigation, we reviewed surveillance data from the National surveillance database. We conducted a Knowledge, Attitudes, and Practices (KAP) survey in conjunction with a seroprevalence survey in the 12 rural villages that reported at least one CCHF case each in 2014. Review of surveillance records revealed that eighteen (82%) case-patients resided in rural villages and 14 (64%) reported a tick exposure. We visited 457 randomly selected households during the one week of data collection. We conducted 616 interviews and obtained 448 blood specimens. Data entry and serological testing are currently ongoing.

BACKGROUND

Crimean-Congo hemorrhagic fever (CCHF) is a tick-borne zoonotic viral disease of the *Bunyaviridae* family. CCHF is endemic in Africa, Asia, Eastern Europe and the Middle East (1). The principal vector responsible for viral transmission is the *Hyalomma* tick (2). Transmission occurs from the bite of an infected tick or from crushing an infected tick with bare skin. Secondary transmission has been reported from contact with infected animal blood or tissues, or ingesting unpasteurized milk. Human-to-human transmission can occur from exposure to infected blood or bodily fluids; this is typically reported in healthcare settings (3).

Although animals and ticks do not exhibit clinical signs of infection, human infection results in a severe illness which presents as four clinical phases: incubation, pre-hemorrhagic, hemorrhagic, and convalescence. The asymptomatic incubation period typically varies from 3-7 days, depending on the mode of transmission. The pre-hemorrhagic phase manifests as a nonspecific febrile illness and lasts 4-5 days. The hemorrhagic phase lasts 2-3 days and is marked by rapidly progressing symptoms. During this phase, severe cases develop multi-organ failure and shock, leading to death. The reported case fatality rate has varied from 5% to 60% (1-6). The convalescent phase lasts about 9-10 days and is characterized by alopecia, labile pulse, tachycardia, and lethargy (2).

CCHF's clinical severity, transmissibility and infectiousness are responsible for its categorization by the National Institute of Allergy and Infectious Diseases as a Category A pathogen (7).

Reducing human exposure to ticks and contact with infected animal or human blood and tissues are at the core of preventing CCHF in endemic regions (2, 8). Treatment is generally limited to supportive care; consequently early detection and diagnosis are critical for survival. Ribavirin, an antiviral agent, has demonstrated effectiveness in in-vitro models and observational studies. Although currently no formal recommendations for the use of ribavirin in the treatment of CCHF exist, its use has been traditionally reserved for those cases deemed as "severe" (2, 9, 10).

Since January 2014, 22 cases of CCHF have been reported in Georgia, including three deaths. Almost half the population of Georgia resides in rural regions and most cases occurred in a known herding corridor (11). Though endemic in the region, this is the highest number of cases reported annually since being designated a notifiable disease by NCDC in 2009 (12, 13). In response, an emergent educational campaign was implemented in each village that had a CCHF case in 2014 to increase awareness of CCHF. However, the extent of the outbreak as well as the source, mode of transmission and risk factors surrounding these cases were unknown.

METHODS

PHASE 1: REVIEW OF EXISTING DATA

We conducted a review of all current and past CCHF cases in NCDC's Electronic Disease Surveillance System (EIDSS). Risk factors, when collected, were reviewed for each case-patient and laboratory testing for each case-patient was identified. Working in collaboration with the Laboratory of the Ministry of Agriculture (LMA) and the Georgian National Food Agency (NFA), any cattle or tick samples previously collected from at-risk regions, defined as the villages and immediate areas around the villages that had at least one case of CCHF in 2014, were identified for CCHF testing.

Additionally, we performed a targeted evaluation of the surveillance system by interviewing key stakeholders from NCDC, the R. G. Lugar Center for Public Health Research, which is the major reference laboratory in the country, and Battelle Memorial Institute, a nonprofit organization contracted for public health education. The goal was to elucidate recent modifications of the system that could have altered its sensitivity for case detection.

PHASE 2: FIELD INVESTIGATION

We conducted a Knowledge, Attitudes and Practice (KAP) survey and a CCHF serosurvey in the 12 affected rural villages, defined as villages that had at least one case of CCHF in 2014. Participants could participate in the KAP, the serosurvey, or both. The four towns or suburban areas that reported case-patients were not included as some case-patients had urban residences but a rural exposure.

We calculated a total sample size of 904 participants or 457 households by allocating the sample size for each village proportional to the population size in the Georgia 2002 census and based on the following assumptions (Figure 1):

1) The limit of statistical significance (alpha) is 0.05 (95% confidence interval)
2) The prevalence of CCHF seropositivity is 2.7% with confidence limits (precision) of +/- 5%, based on seroprevalence data reported from endemic countries (4, 14-18).
3) An estimated design effect of 1, assuming minimal household clustering
4) An anticipated response rate of 90%.
1) An adult household size of two.

Using Google Earth™ (Google, Mountain View, CA: Version 7.1.2.2041) satellite imagery of the 12 affected villages, rooftops were enumerated for each village. Using a random number generator, rooftops were then randomly selected.

In the field, team supervisors verified household selections; each previously selected structure was categorized as a household, an abandoned house, a summer house (defined as a house that is only inhabited during the summer months), or not a house (e.g. commercial property, church.). If the assigned rooftop was an abandoned house or a summer house, which required neighbor confirmation, or a structure that was not a house, the house closest to the right was chosen as a replacement. No other replacement was undertaken.

A total of 29 staff participated in the field investigation; 25 were locally employed and the remaining four were CDC employees. They were organized into four teams, each composed of one supervisor, at least one phlebotomist, and 4-6 interviewers. Household visits were conducted by pairs of interviewers.

During the household visits, all adult household members who met the inclusion criteria were identified. Although the goal was to interview at least 2 participants per household, additional participants in the household were interviewed if available. Inclusion criteria for participation were being an adult (≥18 years old) member of the household who could give consent and residing in the household or village for the preceding two months. Exclusion criteria included CCHF symptoms at the time of the interview, age less than 18 years, not having lived in the village or in the household for the preceding two months, and not being able to give consent.

If a person met the inclusion criteria and consented to participation in the study, the KAP survey was administered. The KAP instrument contained questions on recent illnesses, education received regarding CCHF in the last four months, current knowledge of and practice regarding tick handling, removal, and avoidance, and animal slaughtering practices. Households received educational material about preventing CCHF infection at the conclusion of the interview.

For the serosurvey, a sample of 10 ml whole blood was obtained from each willing participant for CCHF serological testing. Samples were centrifuged and serum was separated into aliquots. Serologic testing will be performed at the R. G. Lugar Center for Public Health Research for recent (within the past 4 months) and past (within last 5 years) CCHF infection as demonstrated by anti-CCHF IgM and IgG, respectively (19). Testing will be performed using the commercially available Vector-Best IgM and IgG kits (Vector-Best Company, Vectocrimean-CHF kit, Novosibirsk, Russia).

Available additional aliquots will be stored at R. G. Lugar Center for Public Health Research for up to 2 years for confirmatory testing, if necessary.

Data Management

KAP survey data will be de-identified and entered using EpiInfo™ (CDC, Atlanta, Georgia). Data will be checked for missing values and entry errors. Investigators will randomly select 10% of records entered in the database for review to ensure proper data entry by comparison with the paper questionnaire. The records reviewed will be selected using a random number generator.

Both serologic and KAP survey data will be analyzed using EpiInfo™ and SAS® 9.3 (SAS Institute Inc., Cary, North Carolina).

Ethical Considerations

Written informed consent was obtained in Georgian and Azeri, as appropriate, by an epidemiologist fluent in the language. All participants were informed that they have the right to refuse to participate and that this would not affect their relationship to NCDC, CDC, or affect their ability to receive healthcare in the future. If anyone exhibited CCHF symptoms at the time of the interview, they would be immediately referred to the nearest public health center for medical evaluation.

Identifying information including name and phone numbers were collected during the administration of the KAP survey. These data will only be used to contact participants regarding their CCHF serologic results. This information will be kept separate from the database, which will contain only de-identified data and a unique identifier for each participant.

Paper records are maintained in a secure, locked file at the CDC Georgia Country Office. At the conclusion of the study, the records will be shredded. All electronic files are kept on a password-protected computer and stored in a secure location.

Results

Phase 1: Review of existing data

Review of all 22 case-patients' records revealed a mean patient age of 45 (Range: 4 to 77); 13 (59%) were male. Eighteen (82%) case-patients resided in rural villages and 20 (91%) cases occurred from May to August (Figure 1). The mean incubation period was 4 days (Range: 1 to 17 days). Preceding their illness, 14 (63.6%) reported a tick exposure (defined as a tick bite or tick removal), 1 (5 %) reported an animal blood exposure, and 7 (32%) were unable to identify their exposure.

CCHF was included in the passive, healthcare facility-based surveillance system starting in 2009. On review of the surveillance system, two recent activities were identified. First, in the past two years NCDC and Battelle Memorial Institute have implemented educational campaigns to increase CCHF awareness, targeting district-level physicians. Second, two studies on undifferentiated acute febrile illness were conducted in Georgia. One study initiated active CCHF surveillance at six hospitals in

2013 (13). The other study, initiated in June 2014 and currently ongoing, investigates cases of fever of unknown origin (FUO) with CCHF included in the laboratory panel (20).

PHASE 2: FIELD INVESTIGATION

Village populations ranged from approximately 248 people to about 3000 people (Figure 1). Most houses were made with wood and had metal roofing. Although most property's had a barn, many animals, especially chickens, were allowed to move freely around the village. The availability of electricity was variable per household and per village. Some households had indoor plumbing but most water was obtained from local springs and wells. Although most residents were welcoming and collaborative, residents living closer to the Russian border were more reserved.

During one week, we visited 522 structures: 475 were houses, 27 (5.2%) were summer houses, 64 (12.3%) were abandoned and 92 (17.6%) were not a house. In the 475 houses visited, occupants were not home in 31 (6.8%), and 13 (2.8%) did not meet the inclusion criteria. We conducted a total of 616 interviews (mean of 1.5 participants per household). Of the selected households where a person was present and inclusion criteria met (n=413), participation rate was 98%. We collected 448 blood specimens (mean of one sample per household, range 0 to 4).

Further results are pending on-going data and serological analysis.

DISCUSSION

The results available at this time indicate most case-patients had some exposure to ticks before the onset of illness, a known risk factor (1, 2). Additionally, the occurrence of cases corresponds to the months in which tick activity is expected to increase. Given the rural environment in which most cases occurred, and the practice of animal husbandry and herding, human exposure to animals and ticks is likely a central risk factor for CCHF transmission.

CCHF case detection by the surveillance system may have been stimulated in the recent past by the educational campaigns and recent CCHF and FUO studies. As the surveillance system relies on physician's considering the diagnosis and testing for it, the educational campaign and implementation of active surveillance, which included physician training, could have resulted in increased case detection. Therefore, at this time, it is unclear whether the increase in CCHF cases in 2014 was truly an outbreak or an artifact of improved sensitivity of the surveillance system.

Seroprevalence data and risk factor analysis are pending.

RECOMMENDATIONS

Since the completion of the field investigation, one additional case-patient has been identified, indicating ongoing transmission, and underscoring the importance of continuing close surveillance. Our current recommendation for NCDC is to intensify ongoing CCHF educational campaigns by focusing on 1) preventing tick exposure and encouraging safe tick handling practices, targeting at-risk populations including herders, farmers, and veterinarians, and 2) minimizing contact with

infected animal blood and tissues, targeting slaughterhouse workers, veterinarians, and healthcare workers.

Additionally, given the interaction between human, animal and tick in CCHF transmission, it is imperative that NCDC, LMA, and NFA continue to collaborate through data sharing and having regular meetings to correlate data.

Our final conclusions and recommendations are pending further data analysis.

Future Plans

We will perform data analysis to identify specific risk factors for CCHF transmission and correlate with the serologic data. Additionally, we will evaluate the effect of the recent educational campaign on knowledge, attitudes and practices in the villages.

Once serologic analysis is complete, any participant that has a positive serological test, whether IgG or IgM, will receive educational information regarding the results. This will be provided by NCDC and the utilization of staff and resources at the local public health centers.

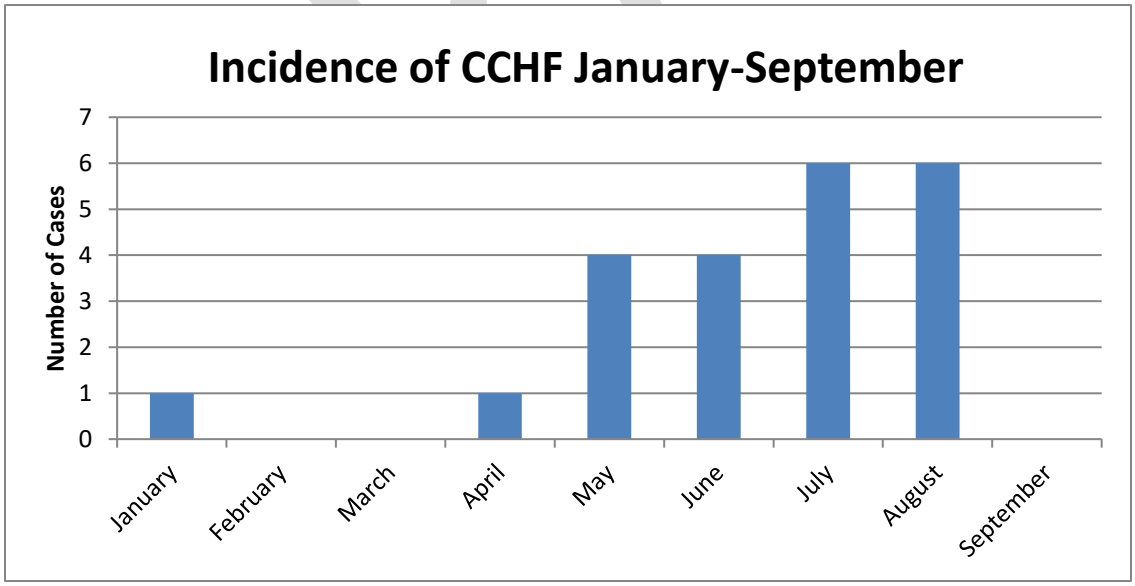
Results and recommendations will be disseminated to local partners through a formal report discussing specific prevention and control recommendations. Additionally, findings will be presented and discussed with key stakeholders to determine the most effective public health actions and messaging.

FIGURES

Figure 1. Breakdown of sample size by each village, Georgia, 2014

	Village Name	Participants	Households	2002 Census Total Population
1	Dviri	71	36	937
2	Zemo Salari	86	43	1135
3	Didi Mejvriskhevi	238.7	120	3153
4	Igoeti	52.4	27	692
5	Bijnisi	28.7	15	379
6	Zekota	18.8	10	248
7	Ali	104.1	53	1375
8	Brili	36	18	475
9	Nabakhtevi	64.8	33	855
10	Vaka	107.8	54	1423
11	Tezeri	59.3	30	783
12	Kemferi	35.6	18	470
	TOTAL	**903.2**	**457**	**11925**

Figure 2. Epidemic Curve of CCHF Cases from January to September, Georgia, 2014



REFERENCES

1. World Health Organization [WHO]. Crimean-Congo haemorrhagic fever: WHO; 2013 [updated January 2013; cited 2014 September 5]. Available from: http://www.who.int/mediacentre/factsheets/fs208/en/.
2. Ergonul O. Crimean-Congo haemorrhagic fever. Lancet Infect Dis. 2006;6:203-14.
3. Appannanavar SB, Mishra B. An update on crimean congo hemorrhagic Fever. Journal of global infectious diseases. 2011;3(3):285-92.
4. Yilmaz GR, Buzgan T, Irmak H, Safran A, Uzun R, Cevik MA, et al. The epidemiology of Crimean-Congo hemorrhagic fever in Turkey, 2002–2007. International Journal of Infectious Diseases. 2009;13(3):380-6.
5. Knust B, Medetov ZB, Kyraubayev KB, Bumburidi Y, Erickson BR, MacNeil A, et al. Crimean-Congo hemorrhagic fever, Kazakhstan, 2009-2010. Emerging infectious diseases. 2012;18(4):643-5.
6. Centers for Disease Control and Prevention [CDC]. Crimean-Congo hemorrhagic fever. 2014 [updated May 9, 2014; cited 2014 September 5]. Available from: http://www.cdc.gov/vhf/crimean-congo/.
7. National Institute of Allergy and Infecitous Diseases [NIAID]. NIAID Biodefense Research [updated August 8, 2014; cited 2014 September 9]. Available from: http://www.niaid.nih.gov/topics/biodefenserelated/biodefense/pages/cata.aspx.
8. Smego RA, Jr., Sarwari AR, Siddiqui AR. Crimean-Congo hemorrhagic fever: prevention and control limitations in a resource-poor country. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2004;38(12):1731-5.
9. Ergonul O, Celikbas A, Dokuzoguz B, Eren S, Baykam N, Esener H. Characteristics of patients with Crimean-Congo hemorrhagic fever in a recent outbreak in Turkey and impact of oral ribavirin therapy. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2004;39(2):284-7.
10. Fisher-Hoch SP, Khan JA, Rehman S, Mirza S, Khurshid M, McCormick JB. Crimean Congo-haemorrhagic fever treated with oral ribavirin. Lancet. 1995;346(8973):472-5.
11. Cental Intelligence Agency. The World Factbook - Georgia [updated June 22, 2014; cited 2014 November 4]. Available from: https://www.cia.gov/library/publications/the-world-factbook/geos/gg.html.
12. Zakhashvili K, Tsertsvadze N, Chikviladze T, Jghenti E, Bekaia M, Kuchuloria T, et al. Crimean-Congo hemorrhagic fever in man, Republic of Georgia, 2009. Emerging infectious diseases. 2010;16(8):1326-8.
13. Kuchuloria T, Imnadze P, Chokheli M, Tsertsvadze T, Endeladze M, Mshvidobadze K, et al. Viral hemorrhagic fever cases in the country of Georgia: Acute Febrile Illness Surveillance Study results. Am J Trop Med Hyg. 2014;91(2):246-8.
14. Sharifi-Mood B, Metanat M, Alavi-Naini R. Prevalence of crimean-congo hemorrhagic Fever among high risk human groups. International journal of high risk behaviors & addiction. 2014;3(1):e11520.
15. Andriamandimby SF, Marianneau P, Rafisandratantsoa JT, Rollin PE, Heraud JM, Tordo N, et al. Crimean-Congo hemorrhagic fever serosurvey in at-risk professionals, Madagascar, 2008 and 2009. Journal of clinical virology : the official publication of the Pan American Society for Clinical Virology. 2011;52(4):370-2.
16. Gergova I, Kamarinchev B. Seroprevalence of Crimean-Congo hemorrhagic fever in southeastern Bulgaria. Japanese journal of infectious diseases. 2014;67(5):397-8.

17. Gergova I, Kamarinchev B. Comparison of the prevalence of Crimean-Congo hemorrhagic fever virus in endemic and non-endemic Bulgarian locations. Journal of vector borne diseases. 2013;50(4):265-70.
18. Sargianou M, Panos G, Tsatsaris A, Gogos C, Papa A. Crimean-Congo hemorrhagic fever: seroprevalence and risk factors among humans in Achaia, western Greece. International journal of infectious diseases : IJID : official publication of the International Society for Infectious Diseases. 2013;17(12):e1160-5.
19. Vanhomwegen J, Alves MJ, Zupanc TA, Bino S, Chinikar S, Karlberg H, et al. Diagnostic assays for Crimean-Congo hemorrhagic fever. Emerging infectious diseases. 2012;18(12):1958-65.
20. Trapaidze N. Interview. US Defense Threat Reduction Agency's Cooperative Biological Research Program in Georgia, GG-21. October 7, 2014.