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Epidemiological and laboratory characterization of a yellow fever outbreak in northern Uganda, October 2010–January 2011

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SUMMARY

Background: In November 2010, following reports of an outbreak of a fatal, febrile, hemorrhagic illness in northern Uganda, the Uganda Ministry of Health established multisector teams to respond to the outbreak.
Methods: This was a case-series investigation in which the response teams conducted epidemiological and laboratory investigations on suspect cases. The cases identified were line-listed and a data analysis was undertaken regularly to guide the outbreak response.
Results: Overall, 181 cases met the yellow fever (YF) suspected case definition; there were 45 deaths (case fatality rate 24.9%). Only 13 (7.5%) of the suspected YF cases were laboratory confirmed, and molecular sequencing revealed 92% homology to the YF virus strain Couma (Ethiopia), East African genotype. Suspected YF cases had fever (100%) and unexplained bleeding (97.8%), but jaundice was rare (11.6%). The overall attack rate was 13 cases/100 000 population, and the attack rate was higher for males than females and increased with age. The index clusters were linked to economic activities undertaken by males around forests.
Conclusions: This was the largest YF outbreak ever reported in Uganda. The wide geographical case dispersion as well as the male and older age preponderance suggests transmission during the outbreak was largely sylvatic and related to occupational activities around forests.

1. Introduction

Yellow fever (YF) is an acute viral hemorrhagic disease caused by the yellow fever virus. The YF virus is an enveloped positive sense single-stranded RNA virus that belongs to the genus *Flavivirus*.[1] The natural host of the virus is non-human primates and the vectors in Africa are usually mosquitoes of species *Aedes africanus* in forest areas and *Aedes aegypti* in urban areas.[2]

In humans, the acute phase occurs 3–6 days after infection and is characterized by non-specific symptoms that last up to 4 days. Fifteen percent of the patients enter a toxic phase characterized by high fever, unexplained bleeding, jaundice, and multiple organ failure, and 50% of these patients die within 2 weeks.[3]

To confirm the disease, serological testing by way of ELISA for YF virus-specific IgM or isolation of the virus from blood samples is usually undertaken, since these are the recommended standard diagnostic tests for YF.[4] Blood samples may be subjected to PCR testing or exceptionally to next generation sequencing (NGS)[5] for the detection of YF virus genetic material. Immunohistochemical techniques are valuable for detecting the viral antigen in liver autopsy tissues.[4] Treatment is supportive since there is no known cure,[6] and vaccination is the mainstay of YF control.[7]

YF is a moving epizootic in endemic regions of tropical Africa and South America lying within a band from 15°N to 10°S of the equator and accounts for an estimated 200 000 cases of YF (with 30 000 deaths) per year globally.[8]

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The largest YF outbreak in East Africa occurred in Ethiopia from 1960 to 1962 and accounted for about 30 000 deaths.[9] YF outbreaks have also been reported from Sudan (1940,[10] 2003,[11] and 2005[12]) and Kenya (1992–93[13]). The first YF outbreak in Uganda was reported in 1941 from Bwamba County, western Uganda.[14] Subsequent YF outbreaks in Uganda were reported in Kabarole District in 1952,[15] Entebbe in 1959 and 1971,[16] and Luwero in 1964.[17]

After 1972, political instability in the country led to a decline in YF surveillance activities in Uganda and hence many suspected YF cases notified by the health facilities were not investigated. Following the introduction of the Integrated Disease Surveillance and Response (IDSR) strategy in Uganda in 2000,[18] a clinical health facility-based surveillance system for YF was instituted, though most of these reports were not accompanied by specimens to facilitate laboratory investigations. It is therefore possible that undetected human cases were occurring despite the absence of confirmed outbreaks in the country over the past four decades.

In November 2010, the Uganda Ministry of Health received reports of an outbreak of a fatal, febrile, hemorrhagic illness in northern Uganda. Multisector teams were established to support the outbreak response. This report highlights the activities and findings of the epidemiology and laboratory team during the outbreak. Entomological and reactive vaccination outcomes will be reported separately.

2. Methods

2.1. Epidemic site

A total of 15 districts in northern Uganda, including Abim, Agago, Apac, Kitgum, Kaabong, Kotido, Lamwo, Arua, Lira, Pader, Gulu, Nebbi, Napak, Dokolo, and Yumbe reported suspected cases of YF (Figure 1). The districts share borders with southern Sudan and Kenya where outbreaks have been reported in the past.[10–13] The northern region is home to a number of game reserves and parks including Kidepo Valley National Park and Murchison Falls National Park. Northern Uganda has just recovered from a nearly two-decade civil war that confined more than 90% of the population to internally displaced people's (IDP) camps. The YF outbreak occurred within 2 years after these people had returned to their original homes.

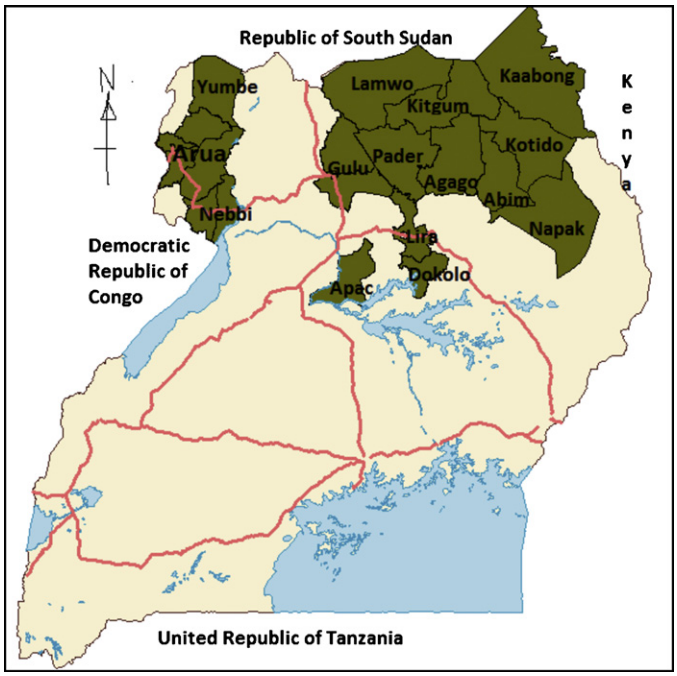

Figure 1. Map showing districts reporting suspect cases, northern Uganda, 2010–2011.

2.2. Investigation teams

The epidemiology and laboratory team was one of the subcommittees of the national task force established to respond to the outbreak. The experts on the team included medical epidemiologists, physicians, laboratory experts, and social workers drawn from the Ministry of Health, other sectors in government, and partner organizations like the World Health Organization (WHO), Centers for Disease Control and Prevention (CDC), Médecins Sans Frontières (MSF), and the African Field Epidemiology Network (AFENET).

2.3. Epidemiology and laboratory activities

2.3.1. Epidemiology

This was a case-series investigation in which the field teams conducted detailed clinical descriptions of all suspected cases. A working case definition was developed for the initially unknown disease, and following the identification of YF using the NGS approach,[19] this was modified (Table 1) to facilitate the identification of additional YF cases at the health facility and community level. All new suspected YF cases were given supportive treatment[4] at designated treatment centers that maintained case line-lists with key variables including: identifiers, age, sex, occupation, residence, date of onset of illness, date of admission to health facility, clinical signs and symptoms, YF vaccination status, types of specimens collected, date of specimen collection, laboratory results, case classification, and case outcome.

2.3.2. Laboratory

Prior to the confirmation of YF, the types of specimens taken reflected the proposed differential diagnoses (Table 2). Hence from each suspected case, one to five blood specimens of 1–3 ml each were obtained for blood culture, viral serology and genetic testing by PCR and/or NGS, blood chemistry, full blood counts, and blood smears. Stool was also obtained in a plain container for microscopy or placed in Cary–Blair transport medium for stool culture. Liver autopsy specimens were obtained and preserved in10% formalin to facilitate YF antigen detection through immunohistochemical testing. After the confirmation of YF by NGS,[19] samples from new suspected cases were collected for blood chemistry, full blood counts, and YF testing by PCR and IgM, with confirmation by plaque reduction neutralization test (PRNT). Because of an ongoing hepatitis E epidemic in northern Uganda, specimens testing negative for YF were subsequently tested for hepatitis E virus and other infections, as elaborated in Table 2.

2.4. Data analysis

A Microsoft Excel data-entry screen incorporating all the variables on the YF line-list was developed and used to capture all the information on the cases reported during the outbreak. The population projections for the affected districts[20] were obtained and used to compute disease attack rates by age, sex, and geographic location. Attack rate maps were drawn using Epi Map software.[21] Epidemic curves were drawn using the dates of onset to determine outbreak trends.

All epidemiological and laboratory data were collected as part of the routine outbreak investigation by national multisector teams, hence ethical clearance was not obtained.

Table 1
Case definitions for the epidemiological investigation of the yellow fever outbreak in northern Uganda, 2010–2011

Period	Classification	Definition
November 8, 2010 to December 30, 2010	Suspected case	Any patient presenting with severe headache with or without fever AND at least three of the following signs or symptoms: • Gastrointestinal illness such as vomiting, hematemesis, watery/bloody diarrhea, dark stools (melena), constipation, foul smelling stools, or • Dizziness, or • General weakness, or • Convulsions, or • Any unexplained bleeding from another site
December 31, 2010 to February 9, 2011	Suspected case	Any person with acute onset of fever, with either a negative laboratory test (blood slide or RDT) for malaria or failure to respond to a full course of antimalarials AND any one of the following: • Jaundice or scleral icterus appearing within 14 days of onset of the first symptoms, or • Unexplained bleeding from either the mouth, nose, gums, skin, eyes, or stomach (gastrointestinal tract)
	Probable case	Any person meeting the suspected case definition criteria WITH either: • An epidemiological link to a confirmed case or the yellow fever outbreak in northern Uganda, or • Positive post-mortem liver histopathology
	Confirmed case	Any person meeting the suspected or probable case definition criteria AND one of the following: • Detection of yellow fever virus-specific IgM • Detection of yellow fever virus-specific neutralizing antibodies • Detection of yellow fever virus genome in blood or liver by PCR or NGS

RDT, rapid diagnostic test for malaria; IgM, immunoglobulin M; PCR, polymerase chain reaction; NGS, next generation sequencing.

3. Results

3.1. Outbreak investigation and response

Following prolonged outbreak investigations that lasted 40 days, laboratory confirmation of YF was made on December 18, 2010 by the CDC (Atlanta, GA, USA) using a random-primed pyrosequencing approach.[19] Consequently, the Ministry of Health declared an outbreak of YF in northern Uganda. A national response plan prioritizing surveillance and laboratory confirmation, case management, social mobilization and health education, and reactive vaccination was developed to control the outbreak.

3.2. Index case investigations

In Abim District, the index case was a 41-year-old male from the village of Wipolo, Aremo Parish, Morulem Sub-County. He frequented the forest to collect bamboo for sale in the local market. This forest is located near the village and most homes are located within a 0.5–1.5 km distance of the forest. His illness started on October 2, 2010 and was characterized by fever, headache, epigastric pain, hematemesis, and melena. He was not treated at any health facility and died 5 days after the onset of illness. He had not traveled out of his home village or stayed with any sick persons prior to the onset of illness. A total of five cases resulting in two deaths were reported from this family, who shared the same house. The other four affected house occupants included a 10-year-old female, a 12-year-old male who died, an eight-year-old male, and a 16-year-old male. None of the four had traveled to the forest, but they went down with a similar illness.

In Kitgum District, the index case was a 38-year-old male hunter from the village of Pudpud, Okuti Parish, Orom Sub-County. The index case village is located at the edge of a forest, and hunting gadgets were recovered from the village. His illness started on November 16, 2010 with complaints of headache, fever, epigastric pain, epistaxis, red eyes, vomiting dark blood, and passing feces with blood. He was not taken to any health facility, but was treated with unspecified medication and died on November 22, 2010. The deceased had neither traveled out of the village nor stayed with sick persons prior to the onset of illness. Three weeks after he died, the patient's mother and son fell sick with similar manifestations. This family had three cases and

Table 2
Differential diagnoses investigated during the yellow fever outbreak in northern Uganda, 2010–2011

Type of illness	Differential diagnoses	Laboratory tests undertaken	Name of Laboratory
Bacterial	Complicated shigellosis	Stool cultures	Central Public Health Laboratories, Uganda; CDC Atlanta, USA
	Typhoid fever	Stool and blood cultures	Central Public Health Laboratories, Uganda; CDC Atlanta, USA
	Escherichia coli (enterohemorrhagic)	Stool cultures and Shiga toxin testing	Central Public Health Laboratories, Uganda; CDC Atlanta, USA
	Campylobacter jejuni	Stool cultures	Central Public Health Laboratories, Uganda; CDC Atlanta, USA
	Yersinia enterocolitica	Stool cultures	Central Public Health Laboratories, Uganda; CDC Atlanta, USA
	Clostridium perfringens	Stool cultures	Central Public Health Laboratories, Uganda; CDC Atlanta, USA
	Anthrax	Rapid tests, blood cultures	Central Public Health Laboratories, Uganda
	Plague	Rapid tests, blood cultures	Uganda Virus Research Institute Plague Laboratory, Arua; CDC Fort Collins, USA
Protozoal	Entamoeba histolytica	Stool microscopy	Central Public Health Laboratories, Uganda
	Severe malaria	Blood slide for malaria parasites	Central Public Health Laboratories, Uganda
Viral	Ebola/Marburg, Lassa fever	Serology (IgM, IgG), PCR	Uganda Virus Research Institute; CDC Atlanta, USA
	Dengue fever	Serology (IgM), PRNT	CDC Dengue Fever Laboratory, Puerto Rico
	West Nile virus	Serology (IgM), PRNT	CDC Dengue Fever Laboratory, Puerto Rico
	Yellow fever	Serology (IgM), PRNT	Uganda Virus Research Institute; CDC Atlanta, USA
	Rift valley fever	Serology (IgM, IgG), PCR	Uganda Virus Research Institute; CDC Atlanta, USA
	Fulminant acute viral hepatitis E	Serology (IgM), PCR	Uganda Virus Research Institute
Intoxication	Adulterated alcohol	Chemical assay (methanol)	Uganda Government Analytical Laboratory

CDC, Centers for Disease Control and Prevention; IgM, immunoglobulin M; IgG, immunoglobulin G; PCR, polymerase chain reaction; PRNT, plaque reduction neutralization test.

one death. The index case in Kitgum was not epidemiologically linked to the index case in Abim.

3.3. Epidemiological description of cases

A total of 273 suspected YF cases including 58 deaths were reported during the period October 2, 2010 to January 28, 2011 from the 15 districts in northern Uganda. The districts included Abim, Agago, Apac, Kitgum, Kaabong, Kotido, Lamwo, Arua, Lira, Pader, Gulu, Nebbi, Napak, Dokolo, and Yumbe.

However, case-based information was available for only 250 (91.6%) suspected cases and 55 (94.8%) deaths that met the initial (unknown disease) case definition. Among the cases with case-based information who met the initial case definition, 181 suspected YF cases presented with fever and unexplained bleeding or jaundice and hence met the second YF case definition; of these cases, 45 died (case fatality rate (CFR) 24.9%). The subsequent analysis therefore applies to the 181 cases and 45 deaths that met the case definition for YF.

The CFR among males (29.6%; 32/108) was nearly twice that of females (17.8%; 13/73). The cases meeting the case definition for suspected YF were reported from the 10 districts of Abim (29 cases), Agago (41 cases), Apac (one case), Gulu (three cases), Kaabong (49 cases), Kitgum (35 cases), Lamwo (14 cases), Napak (two cases), Nebbi (one case), and Pader (six cases).

Among the 181 cases that met the case definition for suspected YF, 173 (96%) underwent laboratory testing for YF, and only 13 (7.5%) were laboratory confirmed as YF; these cases came from five (50%) of the districts with cases meeting the definition for suspected YF. These districts included Agago (two cases), Abim (seven cases), Kitgum (two cases), Pader (one case), and Lamwo (one case). The CFR among YF confirmed cases was 53.8% (7/13).

The age of YF suspected cases varied from 3 months to 83 years, with a mean of 28.2 years, a standard deviation of 17.5 years, and an interquartile range of 24 years. The age among patients was almost normally distributed, with a slight skewing towards the older age groups.

3.4. Clinical presentation

Suspected YF cases presented with fever (100%), any form of unexplained bleeding (97.8%), headache (71.3%), non-bloody vomiting (59.7%), hematemesis (52.5%), epistaxis (42.0%), bloody stools (40.3%), bleeding from at least two sites (13.3%), and jaundice (11.6%).

The duration between illness onset and recovery varied from 3 to 37 days, with a median of 6 days, a mean of 9 days, and a standard deviation of 8.9 days. Amongst patients who died, the duration between onset of illness and death varied from zero to 21 days, with a median of 3 days, a mean of 4 days, and a standard deviation of 5 days.

The initial case occurred in the 39th epidemiological week of 2010 in Abim District (Figure 2). Epidemiological investigations were initiated in the 45th epidemiological week of 2010 after reports of a febrile hemorrhagic illness emerged in Abim District. In the subsequent weeks there was a gradual increase in cases reported from the 10 affected districts, reaching a plateau that stretched from the 46th to the 50th epidemiological week of 2010; the number of reported cases declined from the 51st epidemiological week of 2010 to the 6th epidemiological week of 2011. Laboratory confirmation was made in the 51st epidemiological week of 2010, a delay of 6.5 weeks after investigations commenced. Reactive vaccination was initiated in the 3rd epidemiological week of 2011.

The overall attack rate (cases per 100 000 population) for YF in the five districts where an outbreak was confirmed was 13; this

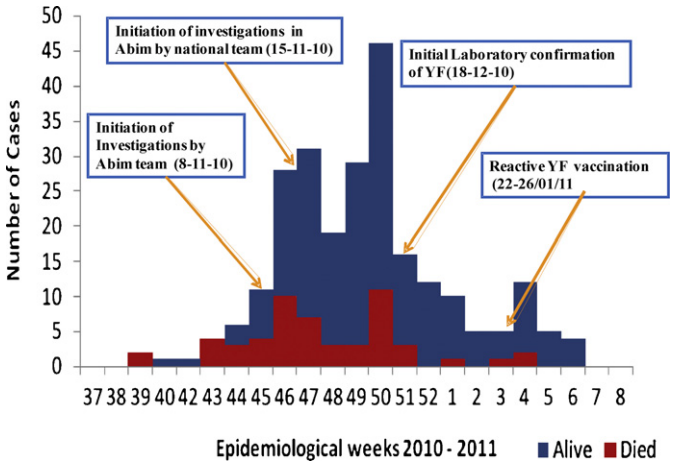

Figure 2. Suspect yellow fever cases by week of onset, northern Uganda, 2010–2011.

varied from 2.9 in Pader to 32.1 in Abim District (Table 3). The risk of YF transmission was highest in Morulem Sub-County in Abim District and Paimol Sub-County in Agago District (Figure 3).

In the five districts where YF was confirmed, the sex-specific attack rates showed that males (16.5/100 000 population) were more affected than females (9.6/10 000 population), with the risk being at least twice as high in males compared to females in the districts of Abim, Agago, and Lamwo (Table 4). In addition, the risk of YF infection increased with age (Figure 4).

By occupation, the cases in the five districts with confirmed cases of YF included peasant farmers (36.5%), children (17.3%), housewives (13.5%), security forces (5.8%), hunters (3%), and borehole drillers (1%).

3.5. Laboratory findings

More than half of the patients had a low hemoglobin level (50.8%); 46.0% had low platelets, 41.3% had lymphocytosis, 38.1% had neutropenia, and 15.9% had leukopenia. Leukopenia with lymphocytosis was identified in 12.7% of cases (Table 5). Liver transaminases were raised in 30.2% of cases for aspartate aminotransferase (AST) and 7.9% of cases for alanine aminotransferase (ALT).

Out of 173 specimens tested, only 13 were confirmed positive for YF: four cases by PCR, eight cases by IgM and PRNT, and one case by NGS (454) testing. Molecular sequencing in one of the confirmed cases revealed 92% homology to the YF virus strain Couma (Ethiopia), belonging to the East African genotype.[19] Two cases from Agago District were flavivirus-indeterminate following PRNT testing. Of the samples from Kaabong District, 17.9% (5/28) were positive for hepatitis E virus-specific IgM and/or PCR. Seven patients were positive for plague using a non-validated rapid antigen test. However, all confirmatory tests for plague (i.e., blood culture, direct fluorescent antibody test, and phage lysis) were

Table 3
Yellow fever suspected case distribution by district, northern Uganda, 2010–2011

District	Alive	Dead	Total	District population 2010	Attack rate (cases/100 000)
Abim	16	13	29	90 306	32.1
Agago	29	12	41	271 700	15.1
Kitgum	28	7	35	254 800	13.7
Lamwo	11	3	14	132 300	10.6
Pader	3	3	6	210 100	2.9
Total	87	38	125	959 206	13.0

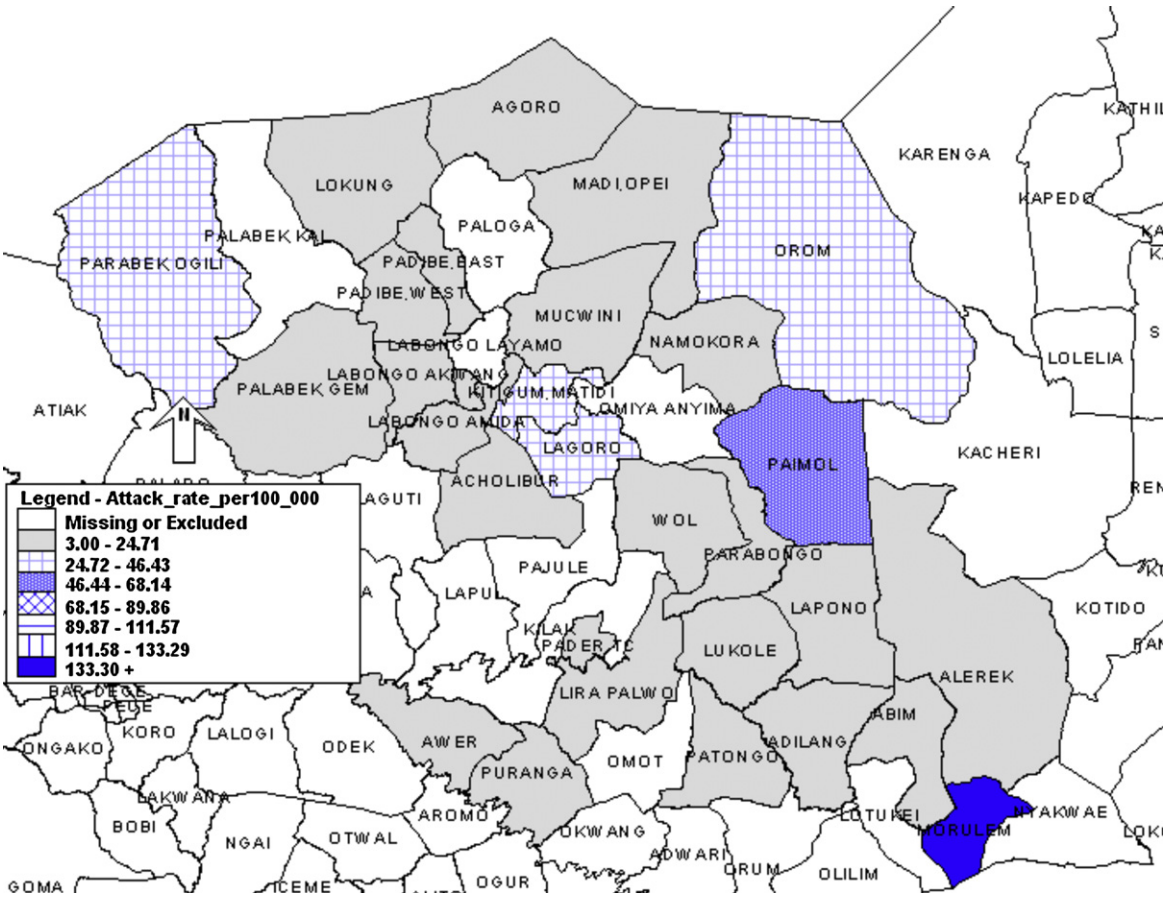

Figure 3. Distribution of suspect yellow fever cases by sub-county, northern Uganda, 2010–2011.

Table 4
Distribution of yellow fever suspect cases by sex, northern Uganda, 2010–2011

District	Male	Female	Male population 2010	Female population 2010	Attack rate in males (cases/100 000)	Attack rate in females (cases/100 000)
Abim	20	9	44 250	46 056	45.2	19.5
Agago	28	13	135 500	136 200	20.7	9.5
Kitgum	19	16	126 500	128 300	15.0	12.5
Lamwo	10	4	67 300	65 000	14.9	6.2
Pader	2	4	105 700	104 400	1.9	3.8
Total	79	46	479 250	479 956	16.5	9.6

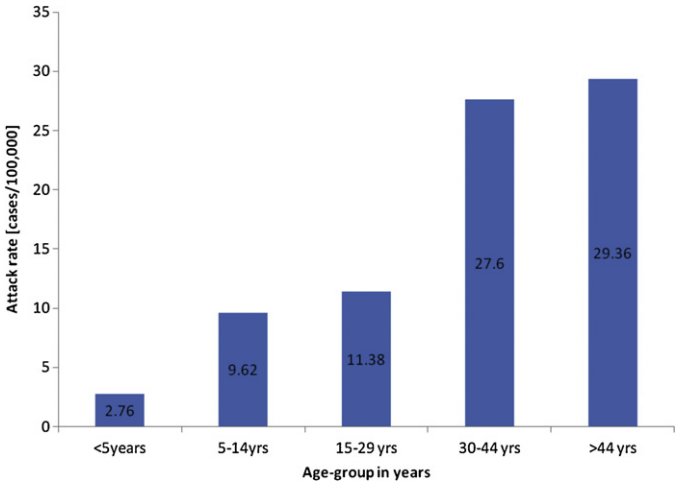

Figure 4. Suspect yellow fever case attack rate by age-group in the districts with confirmed cases, northern Uganda, 2010–2011.

negative. One sample from a suspected case in a 62-year-old female from the neighboring district of Gulu had a positive IgM serological test for Rift Valley fever. Additional testing was undertaken on samples for the differential diagnoses listed in Table 2, but test results were negative for those diseases.

4. Discussion

During the period October 2010 to January 2011, Uganda experienced an outbreak of YF in the north, the largest ever recorded in the country. The overall attack rate in the five districts with confirmed cases was 13 cases per 100 000 population. Prior to the outbreak in northern Uganda, all of the previous five YF outbreaks in Uganda had fewer[14] or single cases.[15–17] However, larger outbreaks have been reported in East Africa, with attack rates (cases per 100 000 persons) of 10 000 in Ethiopia,[9] 6800 in Sudan,[10] and 27.4 in Kenya.[13]

In terms of outbreak severity, the CFR of 53.8% reported among YF confirmed cases in the northern Uganda outbreak is higher than that of Ethiopia (30%),[9] Sudan (10%),[10] and Kenya (19%).[13] The CFR

Table 5
Summary of interpretations from basic investigations on suspected yellow fever cases, northern Uganda, 2010–2011

Parameter	Frequency ($N = 63$)	Percentage
Low hemoglobin	32	50.8
Low platelets	29	46.0
Lymphocytosis	26	41.3
Neutropenia	24	38.1
Raised AST	19	30.2
Leukopenia	10	15.9
Raised creatinine	9	14.3
Lymphopenia	9	14.3
Leukopenia with lymphocytosis	8	12.7
Raised ALT	5	7.9
Raised ALT and AST	5	7.9
Raised total bilirubin	5	7.9
Raised hematocrit	3	4.8
Leukocytosis	3	4.8
Neutrophilia	2	3.2
Raised urea	1	1.6
Raised creatinine and urea	1	1.6
Leukocytosis and neutrophilia	1	1.6

AST, aspartate aminotransferase; ALT, alanine aminotransferase.

also measures the quality of clinical care, which is premised on a timely and definitive diagnosis. During the YF outbreak in northern Uganda, laboratory confirmation did not occur until 40 days after investigations were initiated and hence there was a delay in initiating the recommended supportive treatment[6] and reactive vaccination. The delay in obtaining laboratory confirmation during the YF outbreak in northern Uganda is attributable to several factors. The atypical presentation of cases was one of the reasons for the delayed confirmation, together with the observation by the physicians that patients improved on antibiotics and supportive treatment. The patients identified during the YF outbreak in northern Uganda presented with a febrile hemorrhagic illness that was characterized by fever (100%), any form of unexplained bleeding (97.8%), and headache (71.3%). Jaundice was a rare manifestation and was only reported in 11.6% of suspected YF cases. Hence, given the country's recent experience with filovirus hemorrhagic fever outbreaks,[22,23] Ebola and Marburg were the top diseases on the list of differential diagnoses (Table 2), resulting in these two diseases being prioritized for laboratory testing. The filovirus tests were, however, negative even after repeat testing was undertaken at both the Uganda Virus Research Institute (UVRI) and CDC Atlanta. Additionally, once YF was confirmed by NGS testing,[19] specimens had to be shipped out of the country since testing for YF was not available in Uganda, and this contributed to delayed confirmation of suspected cases. Due to the weak, case-based, laboratory-backed surveillance for YF in the country in recent years, there was no capacity for YF testing in-country when the outbreak started, hence the need to refer specimens to an international laboratory.

An investigation into the index clusters in the three districts of Abim, Agago, and Kitgum revealed that the presentation of cases was consistent with a febrile hemorrhagic illness with jaundice as a rare manifestation. However, despite the initial suspicion of a filovirus outbreak, there was inconsistent evidence of person-to-person transmission among close contacts of suspected or confirmed cases. The epidemic curve for the YF outbreak in northern Uganda showed a gradual rise in cases before reaching a plateau that lasted for close to 4 weeks, indicating a continuous common-source epidemic, a pattern that is seen in vector-borne disease outbreaks like YF due to delayed initiation of reactive vaccination. The case definition of YF that emphasizes jaundice needs to be reviewed in line with the case presentation in this outbreak to avoid missing YF cases in routine case-based surveillance.

During the outbreak, males were more affected than females and the risk of infection increased with age. In Africa, humans are seasonally exposed to YF and hence children who lack naturally acquired immunity are at high risk of disease.[15] Serologic testing among suspected patients during the YF outbreak in northern Uganda suggests limited natural or vaccine-induced immunity, hence adult males who undertake activities inside or close to forested areas were at higher risk of infection prior to the reactive vaccination campaign. It therefore seems likely that most human cases during the outbreak were probably infected by sylvatic vectors. Additionally, until 2 years prior to the outbreak, the civil strife in the region had confined families to IDP camps. At the time the outbreak occurred, most of the families had just returned to their original homes and hence had to clear forests and/or bush after being away for close to two decades. In view of the fact that there were at least two suspected YF cases in each of the case-series without evidence of travel to forests, this raises the possibility of urban transmission during the outbreak.

A limitation of this study arises from the fact that not all suspected YF cases underwent laboratory investigation. The few suspected cases of YF that were not tested could easily have had any of the diseases listed as differentials, particularly hepatitis E virus infection, which has been rampant in the region since 2007.[24] Also, 92.8% of suspected YF cases were negative for YF, indicating that the suspected case definition was very sensitive; this may be attributable to the heightened public and clinical concern regarding the unknown illness and an initially sensitive working case definition that was subsequently made specific for YF, but still included non-specific signs like epistaxis, which captured suspected cases who did not have YF. This may be a reflection of the hysteria created in the country by this outbreak. Many cases of febrile illness with any signs of bleeding were reported as suspected YF cases. Most of these were not confirmed as YF cases and were not investigated further. The high CFR in non-YF cases deserves further investigations.

Following the re-emergence of YF in Uganda, we recommend that case-based laboratory-backed surveillance for YF is strengthened. YF should be considered as a differential in all cases presenting with a febrile hemorrhagic illness, even in the absence of jaundice. In addition, the in-country capacity for YF testing should be re-established. A nationwide risk assessment should be undertaken to inform the national YF control strategy.

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Conflicts of interest: There were no potential conflicts of interest tendered by any of the authors.

References

1. Schoub BD, Venter M. Flaviviruses. In: Zuckerman AJ, Banatvala JE, Schoub B, Griffiths PD, Mortimer P, editors. *Principles and practice of clinical virology*. 6th ed., Chichester: Wiley-Blackwell; 2009. p. 669–98.
2. World Health Organization. District guidelines for yellow fever surveillance. Geneva: WHO; 1998, Available at: http://www.who.int/vaccines-documents/DocsPDF/www9834.pdf (accessed April 16, 2011).
3. World Health Organization. Yellow fever fact sheet. Fact sheet number 100. Geneva: WHO; 2011, Available at: http://www.who.int/mediacentre/factsheets/fs100/en/ (accessed October 11, 2011).
4. Pan American Health Organization. Control of yellow fever. Field guide. Scientific and Technical Publication No. 603. Washington: PAHO/WHO; 2005. Available at: http://www.paho.org/english/ad/fch/im/fieldguide_yellowfever.pdf (accessed December 28, 2010).
5. Su Z, Ning B, Fang H, Hong H, Perkins R, Tong W, et al. Next-generation sequencing and its applications in molecular diagnostics. *Expert Rev Mol Diagn* 2011;**11**:333–43.
6. Monath PT. Yellow fever: an update. *Lancet Infect Dis* 2001;**1**:11–20.
7. World Health Organization. Yellow fever 1996–1997. *Wkly Epidemiol Rec* 1998;**73**:370–2.
8. World Health Organization. Yellow fever fact sheet. Fact sheet number 100. Geneva: WHO; 2001, Available at: https://apps.who.int/inf-fs/en/fact100.html (accessed October 11, 2011).
9. Serie C, Andral L, Poirier A, Lindrec A, Neri P. Studies on yellow fever in Ethiopia. 6. Epidemiologic study. *Bull World Health Organ* 1968;**38**:879–84.
10. Epidemiology of yellow fever. Br Med J 1948; 1: 1241–2.
11. Onyango CO, Ofula VO, Sang RC, Konongoi SL, Sow A, DeCock KM, et al. Yellow fever outbreak, Imatong, southern Sudan. *Emerg Infect Dis* 2004;**10**:1063–8.
12. World Health Organization. Emergency response to the yellow fever outbreak in Sudan. Geneva: WHO; 2005 , Available at: http://www.who.int/hac/donorinfo/reports/donor_rep_sudan_yellow_fever_november2005.pdf (accessed June 30, 2011).
13. Sanders EJ, Marfin AA, Tukei PM, Kuria G, Ademba G, Agata NN, et al. First recorded outbreak of yellow fever in Kenya, 1992–1993. *Am J Trop Med Hyg* 1998;**59**:644–9.
14. Mahaffy AF, Smithburn KC, Jacobs HR, Gillet JD. Yellow fever in western Uganda. *Trans R Soc Trop Med Hyg* 1942;**36**:9–20.
15. Ross RW, Haddow AJ, Raper AB, Trowell HC. A fatal case of yellow fever in a European in Uganda. *East Afr Med J* 1953;**30**:1–11.
16. EAVRI. East African Virus Research Institute annual report, 1971. East African Community. Entebbe, Uganda: Govt. Printers; 1971 (unpublished).
17. Tulloch JA, Patel KM. Yellow fever in central Uganda, 1964. II. Report of a fatal case. *Trans R Soc Trop Med Hyg* 1965;**59**:441–3.
18. Centers for Disease Control Prevention. Assessment of infectious disease surveillance—Uganda, 2000. *MMWR Morb Mortal Wkly Rep* 2000;**49**:687–91.
19. McMullan LK, Frace M, Sammons SA, Shoemaker T, Balinandi S, Wamala JF, et al. Using next generation sequencing to identify yellow fever virus in Uganda. *Virology* 2012;**422**:1–5.
20. Uganda National Bureau of Statistics. Population projections for 2010 and 2011. Kampala: Uganda National Bureau of Statistics; 2011 (unpublished).
21. Centers for Disease Control and Prevention. Epi Info database and statistics software for public health professionals conducting outbreak investigations, managing databases for public health surveillance and other tasks, and general database and statistics applications. Atlanta, GA: CDC; 2010. Available at: http://www.cdc.gov/epiinfo/ (accessed December 28, 2010).
22. Okware SI, Omaswa FG, Zaramba S, Opio A, Lutwama JJ, Kamugisha J, et al. An outbreak of Ebola in Uganda. *Trop Med Int Health* 2002;**7**:1068–75.
23. Wamala JF, Lukwago L, Malimbo M, Nguku P, Yoti Z, Musenero M, et al. Ebola hemorrhagic fever associated with novel virus strain, Uganda, 2007–2008. *Emerg Infect Dis* 2010;**16**:1087–92.
24. Teshale HE, Grytdal PS, Howard C, Barry V, Kamili S, Drobeniuc J, et al. Evidence of person-to-person transmission of hepatitis E virus during a large outbreak in northern Uganda. *Clin Infect Dis* 2010;**50**:1006–10.