Long-term sequelae after Ebola virus disease in Bundibugyo, Uganda: a retrospective cohort study

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Summary

Background **The limited data available for long-term Ebola virus disease health outcomes suggest that sequelae persist for longer than 1 year after infection. The magnitude of the present outbreak in west Africa necessitates a more complete understanding of the health effects and future medical needs of these patients.**

Methods **We invited adult survivors of the 2007 Bundibugyo Ebola virus outbreak in Uganda and their contacts to take part in an observational study roughly 29 months after the outbreak. We collected information about health status, functional limitations, and demographics. We collected blood samples for clinical chemistry, haematology, and filovirus antibodies using ELISA. Analyses were restricted to probable and confirmed survivors and their seronegative contacts.**

Findings **We recruited 70 survivors of the 2007 Bundibugyo Ebola virus and 223 contacts. We did analyses for 49 probable and confirmed survivors and 157 seronegative contacts. Survivors of the Bundibugyo Ebola virus were at significantly increased risk of ocular deficits (retro-orbital pain [RR 4·3, 95% CI 1·9–9·6; p<0·0001], blurred vision [1·9, 1·1–3·2; p=0·018]), hearing loss (2·3, 1·2–4·5; p=0·010), difficulty swallowing (2·1, 1·1–3·9; p=0·017), difficulty sleeping (1·9, 1·3–2·8; p=0·001), arthralgias (2·0, 1·1–3·6; p=0·020), and various constitutional symptoms controlling for age and sex. Chronic health problems (prevalence ratio [PR] 2·1, 95% CI 1·2–3·6; p=0·008) and limitations due to memory loss or confusion (PR 5·8, 1·5–22·4; p=0·010) were also reported more frequently by survivors of Bundibugyo Ebola virus.**

Interpretation **Long-term sequelae persist for more than 2 years after Ebola virus disease. Definition of health consequences related to Ebola virus disease could improve patient care for survivors and contribute to understanding of disease pathogenesis.**

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Introduction

Ebola virus outbreaks have been reported with increasing frequency since 2000.[1] Currently, west Africa is experiencing the largest Ebola virus disease outbreak in history, with more cases recorded in this epidemic than in all other outbreaks combined. Five distinct species in the Ebola virus genus exist, four of which are aetiological agents of Ebola virus disease: Zaire Ebola virus, Sudan Ebola virus, Taï Forest Ebola virus, and Bundibugyo Ebola virus. The case to fatality ratio of Ebola virus disease varies, ranging from roughly 25% for Bundibugyo Ebola virus to 60–90% for Zaire Ebola virus, although survival can be improved with appropriate intensive care.[2,3] The present west African epidemic has already resulted in thousands of survivors, necessitating a more complete understanding of the long-term health effects and future medical needs of these patients.

Severe disease, such as Ebola, is thought to put survivors at increased risk of future adverse health events. The long-term health consequences of Ebola virus infection have not been rigorously assessed. Clinical manifestations arising during convalescence from Ebola virus disease (about 2 weeks to 2 months after disease onset) have been reported, including arthralgias in the large joints, vision loss or uveitis, orchitis, and hearing loss.[4] Anecdotal reports note various persistent sequelae after 1 year, such as abdominal pain, hearing loss, ocular deficits, bleeding, psychological problems, and general malaise.[5] A prospective study by Rowe and colleagues[6] followed up survivors of the Zaire Ebola virus outbreak in Kikwit, Democratic Republic of the Congo for up to 21 months and reported arthralgias and myalgia were significantly more frequent in the 29 Zaire Ebola virus survivors compared with 152 household contacts (62% *vs* 3·8% for arthralgias and 47% *vs* 3·7% for myalgia).[6] Kibadi and colleagues[7] undertook focused ocular examinations of four survivors of the Kikwit outbreak with symptoms including ocular pain, photophobia, hyperlacrimation, and loss of visual acuity. All four patients were diagnosed with uveitis, and symptoms resolved upon treatment with topical 1% atropine and steroids.

Additionally, data from previous reports suggest functional limitations of Ebola virus disease survivors. Most survivors of the Sudan Ebola virus outbreak in Gulu, Uganda were reportedly unable to perform their previous work up to 1 year after infection, with obvious economic consequences.[5] 70% of the survivors of the Zaire Ebola virus outbreak in Kikwit indicated that their capacity to work was worse at the 21-month follow-up than before their infection.[6] Gaining further insight into the

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Research in context

Evidence before this study

Few studies have rigorously assessed the long-term health consequences of Ebola virus infection. In October, 2014, we searched PubMed and Medline for studies investigating sequelae in survivors of Ebola virus disease published in any language. Because the scientific literature on this topic is sparse, we did not restrict our search based on language, study design, geographic location, or patient characteristics. We included keyword searches using Boolean operators and the following search terms: "ebola", "Ebola virus", "survivors", "sequelae", "chronic", and "convalescent".

Added value of this study

Our results add to the small body of scientific literature suggesting that survivors of Ebola virus infection have chronic sequelae including ocular deficits, hearing loss, sleep disturbances, arthralgias, and various constitutional symptoms. Our findings are novel because we noted that these sequelae persisted for longer than 2 years after infection. Furthermore, the long-term health consequences of Bundibugyo Ebola virus infection have not been previously reported.

Implications of all the available evidence

The continuing Ebola virus disease outbreak in west Africa has resulted in thousands of deaths, but also thousands of survivors. Our findings and those of other studies suggest that strategies to address the long-term health needs of survivors are needed.

characteristics and magnitude of the impairment caused by Ebola virus disease might help with the development of preventive and therapeutic approaches.

We aimed to establish the long-term clinical manifestations and limitations in physical function resulting from the outbreak of Bundibugyo Ebola virus in the Bundibugyo District of Uganda in 2007. This outbreak resulted in roughly 192 suspected cases, 116 of whom were later classified as probable or confirmed cases, and 39 deaths.[5,8–10]

Methods

Study design

In this retrospective cohort study, we assessed the long-term health outcomes potentially resulting from clinical Ebola virus disease. The protocol was approved by institutional review boards at the Makerere University School of Public Health, Kampala, Uganda, and the Walter Reed Army Institute of Research, Silver Spring, MD, USA. All study participants provided written informed consent. Individuals with the exposure of interest (ie, history of Ebola virus disease) were identified with a list of suspected, probable, and confirmed cases of Bundibugyo Ebola virus from the outbreak in Bundibugyo District that happened between August, 2007, and January, 2008. The list of cases and their contacts was provided by the Epidemiology and Surveillance Division, Ministry of Health of Uganda. The panel shows the case definitions used by the Ministry of Health during the outbreak. A survivor was requested to identify their contacts in cases for which no contacts were listed on the Ministry of Health list.

We contacted survivors of Bundibugyo Ebola virus who were older than 18 years and invited them to participate (figure). We recruited adult contacts of survivors to provide a comparison cohort of unexposed individuals with similar genetic, socioeconomic, and environmental composition to the Bundibugyo Ebola virus survivor cohort. To reduce potential misclassification of exposure status, we restricted the primary analyses to participants classified as confirmed or probable cases at the time of the initial outbreak and their seronegative contacts. We did two sensitivity analyses to assess the robustness of the incident health outcome findings; the first compared confirmed and probable cases (n=49) with all seronegative contacts (n=208), and the second compared seropositive cases (n=32) with all seronegative contacts (n=208).

Data collection

A standardised questionnaire, phlebotomy, and physical exam were administered in private to consenting volunteers by trained study clinicians. Interviews were conducted in the volunteer's native language by interviewers fluent in the language (English, Lwamba, or Rukonzo). Volunteers were informed that they would not receive direct benefit from participating, other than the

Panel: Case definitions used by the Ugandan Ministry of Health during the Bundibugyo Ebola virus outbreak[10,11]

Suspected case

One of the following:

- Resident of or visitor to the affected sub-counties in Bundibugyo District
- Sudden onset of fever with at least four of the following symptoms (since Aug 1, 2007): vomiting, diarrhoea, abdominal pain, conjunctivitis, skin rash, unexplained bleeding from any body part, muscle pain, intense fatigue, difficulty swallowing, difficulty breathing, hiccups, or headache
- Sudden onset of fever with contact with a suspected, probable, or confirmed case

Probable case

- Meets the suspected case definition
- At least three of the following symptoms; vomiting, diarrhoea, or unexplained bleeding from any site, conjunctivitis, or skin rash
- An epidemiological link to a probable or confirmed case, or a suspected case in whom no specimen was collected or had a negative laboratory result but specimen was collected 0–3 days after onset of symptoms

Confirmed case

- Meets the suspected or probable case definition
- Laboratory confirmation of infection by PCR, virus isolation, antigen detection ELISA, immunohistochemistry, or antibody detection ELISA

potential benefits of the physical examination and blood tests. Information was collected about recent health status, history of symptoms arising at the time of the Bundibugyo Ebola virus outbreak, history of severe illness, and current functional level (appendix). Collection of data and clinical samples was started roughly 2·5 years after the outbreak began and was done at one timepoint.

Questions pertaining to the volunteers' recent health status were designed to capture symptoms experienced from the time of the Bundibugyo Ebola virus outbreak until the time of the interview. Volunteers were asked if each symptom was ever experienced from the time after the Ebola outbreak until now, whether the symptom severity was mild, moderate, or severe, and the chronicity (acute, chronic, or episodic). Acute referred to a symptom with a sudden onset and short course, chronic referred to a symptom that persists over time, and episodic referred to a symptom that periodically resolves and returns. Queried symptoms included fever, headache, retro-orbital pain, blurred vision, hearing loss, difficulty swallowing, sensory changes, swollen glands, shortness of breath, cough, fatigue, depressed mood, difficulty sleeping, pain in the joints, stiff joints, muscle soreness, muscle weakness, diarrhoea, abdominal pain, weight loss, unusual bleeding, and impotence.

Symptoms arising at the time of acute Ebola virus disease were characterised in terms of presence (yes, no, or unknown), severity (mild, moderate, or severe), and duration in days. In addition to these symptoms, queried symptoms included anorexia, hiccoughs, seizures, jaundice, vomiting, skin rash, abnormal bleeding from puncture sites, bleeding from the gums, bleeding in the eyes, black or bloody stool, bleeding from the nose, and unusual menstrual bleeding. To assess whether severity of Ebola virus disease is associated with the development of long-term sequelae, we investigated whether seizures or melaena during Ebola virus disease were associated with the symptoms reported significantly more frequently by survivors of Bundibugyo Ebola virus.

We assessed limitations in physical function with the National Health and Nutrition Survey (NHANES) Physical Functioning Questionnaire,[12] which was slightly modified to be applicable in Uganda. Modifications included use of the metric system and use of regionally appropriate examples (eg, sweeping instead of vacuuming). Volunteers were asked to respond to a series of questions about current limitations caused by any long-term physical, mental, or emotional problem. Volunteers were instructed not to include limitations due to temporary disorders like pregnancy or broken bones. The response categories included yes, no, or unknown, and questions using a Likert scale indicating whether an activity was done with no difficulty, some difficulty, much difficulty, unable to perform, or not done. The questions focused on current limitations in ability to function, and did not attempt to capture change in ability to function since the outbreak of Bundibugyo Ebola virus.

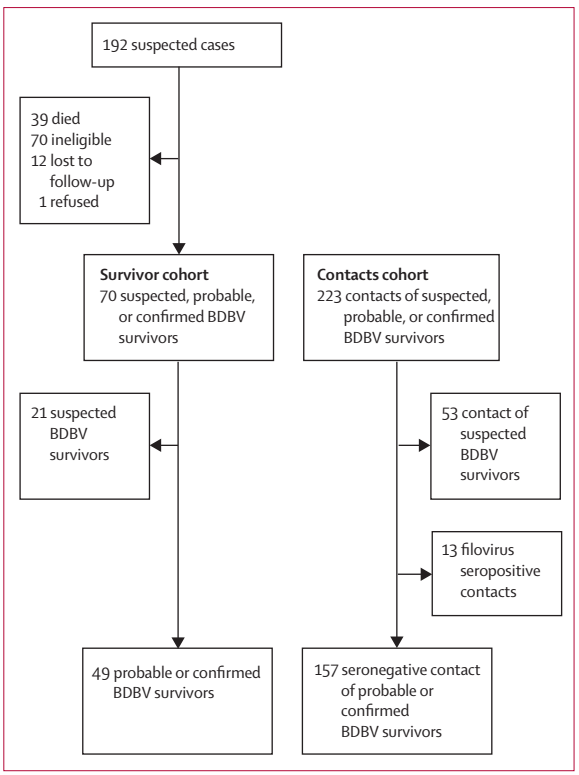

Figure: **Study profile**
Two contacts of suspected Bundibugyo Ebola virus (BDBV) cases had antibodies against filovirus species, for 15 seropositive contacts. We were unable to locate ten survivors, and two survivors had moved to a different district. One survivor refused to participate. Survivors younger than 18 years or those who did not meet the suspect case definition were ineligible for the study.

See **Online** for appendix

Laboratory methods

Serological assays

Serum samples were tested for IgG antibodies against four of the species of Ebola virus, Bundibugyo Ebola virus, Zaire Ebola virus, Sudan Ebola virus, and Taï Forest Ebola virus, and the single Marburg virus species with ELISA as previously described.[13] Plaque reduction neutralisation tests were done on selected sera to confirm the likely specificity of the antibodies to Bundibugyo Ebola virus. Serological testing was done at the US Army Medical Research Institute for Infectious Diseases, Fort Detrick, MD, USA. Samples were processed within 8 h of collection. Sera were cryopreserved at –80°C and shipped on dry ice.

Clinical laboratory assays

Anticoagulated whole blood was used for assessment of complete blood count with a five-part differential on a fully automated platform with the COULTER Ac·T 5diff CP automated haematology analyser (Beckman Coulter, Brea, CA, USA). Serum chemistries (alkaline phosphatase, alanine aminotransfease, aspartate amino-transferase, bilirubin total, blood urea nitrogen, chloride, creatinine, creatinine kinase, gamma-glutamyl

transferase, glucose, lactate dehydrogenase, potassium, and sodium) were done from cryopreserved samples with manufacturer reagents and procedures and run on a COBAS Integra 400 plus automated high-throughput chemistry analyser (Roche Diagnostics, Indianapolis, IN, USA). Cryopreserved sera were also sent to Ebenezer Clinical Laboratory, Kampala, Uganda, for thyroid stimulating hormone (TSH), C-reactive protein (CRP), and Rhesus factor testing. TSH was run on a COBAS e411 automated chemistry analyser (Roche Diagnostics, Indianapolis, IN, USA) with Roche reagent and method. CRP and Rhesus factor were both done on the semi-quantitative agglutination platform with standard reagents.

	Bundi-bugyo	Zaire	Sudan	Täi Forest	Marburg virus, strain Musoke	Any filovirus
Bundibugyo Ebola virus cases (n=70)	32	24	11	12	1	36
Suspected (n=21)	0	2	1	0	0	2
Probable (n=13)	4	2	0	0	1	5
Confirmed (n=36)	28	20	10	12	0	29
Contact (n=223)	8	11	3	1	0	15

Table 1: **ELISA reactivity of patient serum to antigens from four Ebola virus species and Marburg virus**

	Full cohort			Analysis cohort		
	Survivor (n=70)	Uninfected (n=223)	p value	Survivor (n=49)	Uninfected (n=157)	p value
Average age (years)	37·9 (28–46)	33·9 (25–40)	0·02*	40·0 (31–50)	33·5 (24–40)	0·002*
Sex (men)	39 (56%)	107(48%)	0·26	26 (53%)	74 (47%)	0·47
Tribe (Bakhonzo)	60 (86%)	198 (89%)	0·63	43 (88%)	144 (92%)	0·57
Residence (temporary)	4 (6%)	12 (5%)	0·89	1 (2%)	3 (2%)	0·99
Education	..	..	0·02*	..	..	<0·0001*
None	10 (14%)	27 (12%)	..	9 (18%)	13 (8%)	..
Primary	30 (43%)	96 (43%)	..	13 (27%)	65 (41%)	..
Secondary	14 (20%)	77 (35%)	..	12 (25%)	63 (40%)	..
Tertiary	16 (23%)	23 (10%)	..	15 (31%)	16 (10%)	..
Occupation	..	..	0·01*	..	..	0·03*
Farmer	35 (50%)	120 (54%)	..	22 (45%)	80 (51%)	..
Housewife	12 (17%)	42 (19%)	..	6 (12%)	29 (19%)	..
Health worker	11 (16%)	9 (4%)	..	9 (18%)	8 (5%)	..

Data are median (IQR) or n (%). *Significant differences between survivors of Bundibugyo Ebola virus and uninfected participants.

Table 2: **Demographic characteristics of the study population**

	Bundibugyo Ebola virus survivor (n=49)	Uninfected (n=157)	Relative risk* (95% CI); p value
Retro-orbital pain	14 (29%)	8 (5%)	4·3 (1·9–9·6); <0·0001
Muscle weakness	6 (12%)	5 (3%)	3·3 (1·1–10·3); 0·038
Stiffness in joints	11 (22%)	11 (7%)	2·5 (1·1–5·4); 0·022
Hearing loss	13 (27%)	16 (10%)	2·3 (1·2–4·5); 0·010
Difficulty swallowing	13 (27%)	21 (13%)	2·1 (1·1–3·9); 0·017
Joint pain	17 (35%)	18 (11%)	2·0 (1·1–3·6); 0·020
Difficulty sleeping	28 (57%)	41 (26%)	1·9 (1·3–2·8); 0·001
Fatigue	28 (57%)	40 (25%)	1·9 (1·3–2·8); 0·001
Blurred vision	19 (39%)	26 (17%)	1·9 (1·1–3·2); 0·018
Headache	43 (88%)	118 (75%)	1·2 (1·0–1·3); 0·007

Data are n (%). *Relative risk of the outcome in Bundibugyo Ebola virus survivors compared with uninfected participants adjusting for age and sex.

Table 3: **Incident health outcomes in Bundibugyo Ebola virus survivors and uninfected participants**

Statistical analysis

We compared demographic characteristics, clinical data, and clinical laboratory tests between Bundibugyo Ebola virus survivors and uninfected groups with χ^2 or Fisher's exact test, the Student's t-test for normally distributed data, and the Wilcoxon rank sum test for non-normal data. Health status and clinical laboratory comparisons were stratified by sex. The functional relation between continuous variables and health outcome were assessed with LOWESS plots. Log-binomial models were fit to quantify the relative risk of developing the health outcomes for Bundibugyo Ebola virus survivors compared with uninfected participants adjusted for age and sex. In these models, history of Ebola virus disease was modelled as the independent variable. The association of severe Ebola virus disease (reported seizures or melaena) with development of health outcomes was modelled with logistic regression due to convergence failures with the log-binomial models. These models were restricted to confirmed and probable Bundibugyo Ebola virus survivors and adjusted for age and sex. Tests of statistical significance were two-tailed, and significance was defined as $p<0·05$. All analyses were done with Stata (version 11).

Role of the funding source

The funder had no involvement in the writing of the manuscript or the decision to submit for publication. The funder had no role in the study design, data collection, data analysis, or data interpretation. The corresponding author had full access to all of the study data and had final responsibility for the decision to publish the findings.

Results

We enrolled 70 Bundibugyo Ebola virus survivors (classified by the Ministry of Health as suspected, probable, and confirmed cases) and a comparison cohort of 223 contacts (figure). None of the 21 suspected cases had detectable antibodies to Bundibugyo Ebola virus, and only two were reactive to any of the Ebola virus species (table 1). Of the probable cases, four (31%) of 13 had antibodies to Bundibugyo Ebola virus, whereas most (28/36 [78%]) of the confirmed cases had detectable Bundibugyo Ebola virus antibodies. Of the 38 patients who did not have antibodies to Bundibugyo Ebola virus, four had evidence of antibodies to another filovirus. Most survivors with Bundibugyo Ebola virus antibodies also

	Severity (mean)		Acute (%)		Chronic (%)		Episodic (%)	
	Bundibugyo Ebola virus survivor	Uninfected	Bundibugyo Ebola virus survivor	Uninfected	Bundibugyo Ebola virus survivor	Uninfected	Bundibugyo Ebola virus survivor	Uninfected
Retro-orbital pain	1·7	1·6	2 (14%)	6 (75%)	2 (14%)	2 (25%)	10 (71%)	0 (0%)
Muscle weakness	1·4	1·6	3 (60%)	2 (40%)	0 (0%)	0 (0%)	2 (40%)	3 (60%)
Stiffness in joints	1·8	1·7	3 (33%)	2 (29%)	1 (11%)	1 (14%)	5 (56%)	4 (57%)
Hearing loss	1·8	1·6	1 (8%)	6 (40%)	3 (23%)	2 (13%)	9 (69%)	7 (47%)
Difficulty swallowing	1·5	1·8	1 (8%)	5 (26%)	0 (0%)	1 (5%)	11 (92%)	13 (68%)
Joint pain	1·7	1·8	3 (20%)	1 (9%)	1 (7%)	4 (36%)	11 (73%)	6 (55%)
Difficulty sleeping	1·9	2·0	3 (11%)	0 (0%)	6 (22%)	9 (24%)	18 (67%)	28 (76%)
Fatigue	1·8	1·6	5 (18%)	14 (35%)	2 (7%)	3 (8%)	21 (75%)	23 (56%)
Blurred vision	1·6	1·8	2 (11%)	3 (12%)	5 (28%)	12 (46%)	11 (61%)	11 (42%)

Data are n (%).

Table 4: **Severity and chronicity of health outcomes in Bundibugyo Ebola virus survivors and uninfected participants**

had evidence of antibodies to other Ebola virus species, which was expected in view of the known cross-reactivity of IgG antibodies between species.[14] 15 contacts had detectable Ebola virus-specific antibodies; most (13/15 [87%]) were contacts of probable or confirmed cases of Bundibugyo Ebola virus. Two of the 15 seropositive contacts reported no symptoms at the time of the Bundibugyo Ebola virus outbreak; the remaining 13 seropositive contacts reported symptoms including fever (11), headache (11), cough (ten), abdominal pain (five), diarrhoea (three), and unusual bleeding (one). Neutralising antibodies were detectable in 15 survivors and seven contacts.

Table 2 shows the demographic characteristics of the full cohort and the analysis cohort. The analysis cohort was limited to probable or confirmed Bundibugyo Ebola virus survivors and their seronegative contacts to reduce potential misclassification of exposure status. Bundibugyo Ebola virus survivors were older than uninfected participants (40 *vs* 34 years; p=0·002). Not surprisingly, 18% of the Bundibugyo Ebola virus survivors were health workers compared with 5% of uninfected participants (p=0·003).

Bundibugyo Ebola virus survivors were at significantly higher risk for a range of health complaints, adjusting for age and sex (table 3 and appendix). Reported symptoms included ocular deficits, hearing loss, arthralgias, and constitutional symptoms. In particular, the risk of retro-orbital pain was four times higher for Bundibugyo Ebola virus survivors (RR=4·3; p<0·001), and was reported in 29% of survivors compared with 5% of uninfected participants. Survivors with retro-orbital pain mainly reported that the pain was episodic (71%), whereas uninfected participants reported acute retro-orbital pain (75%; table 4). The risk of hearing loss was twice as high for Bundibugyo Ebola virus survivors (RR=2·3; p=0·01), and was reported mainly as moderate loss of hearing by survivors (52%) versus mild hearing loss among uninfected participants (56%). Most survivors (28/49 [57%]) reported fatigue and difficulty sleeping since the time of the outbreak, compared with about 25% (40 and 41, respectively, of 157) of uninfected participants. Survivors and uninfected participants reported fever, swollen glands, shortness of breath, cough, depressed mood, muscle soreness, diarrhoea, abdominal pain, weight loss, unusual bleeding, and impotence with similar frequency. Of Bundibugyo Ebola virus survivors, seizures at the time of the outbreak were associated with development of dysphagia, controlling for age and sex (p=0·038). Melaena during Ebola virus disease was associated with reported joint stiffness among Bundibugyo Ebola virus survivors of similar age and sex (p=0·02).

Both sensitivity analyses gave results of similar magnitude and inference to the primary analyses, with some exceptions (appendix). Depressed mood and weight loss, which were both not significant in the primary analyses (RR 1·9, 95% CI 1·0–3·6; p=0·058 for depressed mood; 1·6, 0·8–3·1; p=0·177 for weight loss), were significant in both the sensitivity analysis comparing confirmed and probable Bundibugyo Ebola virus survivors to all seronegative contacts (2·1, 1·1–4·0; p=0·019 *vs* 2·1, 1·1–4·2; p=0·032) and the sensitivity analysis comparing seropositive survivors with all

	Bundibugyo Ebola virus survivor (n=49)	Uninfected (n=157)	PR (95% CI)	p value
Health problem limits ability to walk or run	14 (29%)	25 (16%)	1·4 (0·9-2·4)	0·165
Health problem lasting ≥12 months	18 (37%)	23 (15%)	2·1 (1·2-3·6)	0·008
Limited in kind or amount of work	1 (2%)	1 (1%)	2·1 (0·1-31·8)	0·602
Limited due to difficulty remembering or confusion	7 (14%)	3 (2%)	5·8 (1·5-22·4)	0·010
Function scale	20·0 (3·5)	18·8 (2·2)	0·77 (–0·0 to 1·6)*	0·055

Data are n (%) or mean (SD). PR=prevalence ratio. *Linear regression controlling for age and sex.

Table 5: **Current limitations in routine function**

seronegative contacts (2·5, 1·3–4·3; p=0·004 *vs* 2·6, 1·3–5·3; p=0·007). Muscle soreness was also significant in the sensitivity analysis comparing seropositive survivors to all seronegative contacts (RR 2·5, 95% CI 1·0–5·9; p=0·040). Hearing loss was reported significantly more in Bundibugyo Ebola virus survivors in the primary analyses (RR 2·3, 95% CI 1·2–4·5; p=0·010), but was no longer significant in the sensitivity analyses comparing seropositive survivors with all seronegative contacts (2·0, 0·8–4·5; p=0·115).

Limitations in the ability to undertake routine functions were more prevalent in survivors, controlling for age and sex (table 5). Specifically, chronic health problems lasting over 12 months were reported more than twice as frequently by Bundibugyo Ebola virus survivors adjusting for age and sex (PR 2·1, 95% CI 1·2–3·6; p=0·008). The chronic health problems reported by Bundibugyo Ebola virus survivors included pain in the abdomen, back, and large joints, fatigue, impotence, and severe headaches. Limitations due to memory problems or confusion were roughly six times more prevalent in Bundibugyo Ebola virus survivors than uninfected participants of similar age and sex (5·8, 95% CI: 1·5–22·4; p=0·010). Health problems resulting in physical inability to walk or run were not significantly different between the two groups. Survivors scored marginally higher (0·77 points) on the physical limitations score adjusting for age and sex (95% CI –0·02 to 1·6, p=0·055), suggesting increased difficulty doing routine functions.

We noted no clinically relevant differences in results from the haematology or clinical chemistry laboratories. Four participants tested positive for HIV (two Bundibugyo Ebola virus survivors and two uninfected participants). Of the two HIV-infected Bundibugyo Ebola virus survivors, only one had a detectable Bundibugyo Ebola virus antibody concentration. General physical exam findings were also not significantly different between Bundibugyo Ebola virus survivors and uninfected participants. However, focused exams such as vision or hearing tests, or neurological exams were not done.

Discussion

Our results show that the risk of developing chronic sequelae is higher for people who survive Ebola virus disease (Bundibugyo Ebola virus) than a control group 2 years after the initial infection. The study by Rowe and colleagues[6] of 29 Zaire Ebola virus survivors in the Democratic Republic of the Congo described sequelae up to 21 months after infection. Those researchers reported an increased incidence of arthralgia and fatigue in survivors similar to our findings. Additional findings from our investigation include hearing loss, ocular deficits, neurological abnormalities, and constitutional symptoms reported more frequently in survivors than uninfected participants. The differences between our two studies suggest that further research is needed to establish whether different species of Ebola virus causing disease impart different risk of long-term sequelae. Our findings provide further evidence supporting anecdotal reports of hearing loss and ocular deficits following Ebola virus disease.[5]

Several infectious diseases cause chronic symptoms that develop or persist after the period of acute infection. Our findings are similar to the post-infectious asthenia described after acute dengue fever.[15] The sequelae we recorded are also analogous to the sensorineural hearing loss recorded in roughly 29% of Lassa virus infections,[16,17] and retinitis or uveitis noted in 1–20% of Rift Valley fever infections.[18] Arthralgia after chikungunya virus infection reportedly persists for 3–5 years in about 10% of patients.[19] Little information is available about the sequelae resulting from infection with Marburg virus, but infection might result in orchitis, hepatitis, or uveitis.[20]

The underlying pathogenesis of post-infectious complications is largely unknown, but is probably multifactorial. Mechanisms could include tissue damage due to direct viral effects, a hypothesis that is supported by results of animal studies showing viral antigen in the brain and eyes of rhesus macaques infected with Zaire Ebola virus.[21] Investigators who studied Zaire Ebola virus survivors in the Democratic Republic of the Congo noted that Zaire Ebola virus specific antibody concentrations were higher in survivors reporting arthralgias than in survivors without arthralgias.[6] Sensineural hearing loss associated with Lassa fever is hypothesised to be either direct viral or autoimmune mediated.[16,17] We did not note raised inflammatory markers in our patient population, although this has been described for other infections with persistent sequelae such as dengue[22] and chikungunya.[23,24]

We detected measureable Bundibugyo Ebola virus antibodies in most survivors recorded as laboratory confirmed cases at the time of the outbreak (78%), suggesting that IgG antibodies to Bundibugyo Ebola virus persist for at least 2 years after infection in more than half of infected individuals. 22% of the confirmed cases had undetectable Bundibugyo Ebola virus antibody concentrations; Bundibugyo Ebola virus induced antibodies probably decreased to undetectable amounts in these survivors, but we cannot exclude the possibility that these patients were misclassified at the time of the original outbreak. We did a sensitivity analysis restricted to seropositive survivors and all seronegative contacts, and our conclusions remain mainly unchanged. Depressed mood, weight loss, and muscle soreness were significant in the sensitivity analyses but not the primary analyses, whereas hearing loss was no longer significant in one sensitivity analysis. Further investigation into the long-term immunological responses to Ebola virus infection is needed to understand protective responses for future prevention and treatment. Additionally, understanding immunological memory responses and associations between Ebola virus antigens and host responses merits further exploration.

Some contacts had detectable IgG antibodies possibly due to crossreactivity to other viruses, asymptomatic infection, or symptomatic infections that were not identified at the time of the outbreak investigation. Previous reports of Ebola virus outbreaks suggest that asymptomatic infection can occur, but infrequently.[6,25] Most seropositive contacts (87%) reported that they experienced symptoms at the time of the Bundibugyo Ebola virus outbreak; some of these individuals might have survived Ebola virus disease. We excluded contacts with detectable Bundibugyo Ebola virus antibody concentrations to reduce potential misclassification of exposure status. Most participants with detectable Bundibugyo Ebola virus antibodies also had detectable antibodies to several other species of Ebola virus. Although some participants might have been previously exposed to Ebola, we expect that this finding is due to antibody cross-reactivity between species.[14]

Some limitations of this investigation should be acknowledged. Measuring subjective symptoms presents inherent limitations. Subjective complications are certainly relevant to a person's health, and are indicative of a personal perception of a decrement in health. However, perceptions of pain and fatigue vary between individuals and populations. We attempted to minimise this issue by introducing scales to quantify subjective complaints. Survivors could have been more keenly aware of their health after the Bundibugyo Ebola virus outbreak, potentially resulting in recall bias. Our use of a comparison cohort of close contacts of Bundibugyo Ebola virus survivors might have minimised this bias because they had witnessed the effects of a severe disease but not experienced it themselves. Use of a comparison cohort with similar genetic, socieconomic, and environmental exposures to the Bundibugyo Ebola virus survivors might also minimise the effect of potential unmeasured confounders, such as drug use or pre-existing comorbidities. Additionally, patient's chronic complications might have been caused by another disorder instead of Ebola virus disease. We tried to minimise the possibility of alternate causality by comparing the results in infected patients with a comparison cohort.

Our assessment of physical function focused on current ability to undertake specific tasks, and thus did not capture changes in ability to function after the outbreak. Although we cannot say that an individual's ability to function diminished after infection with Ebola virus, we can compare their current ability to function with that of unexposed individuals of similar age and sex. The available data did not allow us to account for differences in the time between Bundibugyo Ebola virus infection and enrolment; some Bundibugyo Ebola virus survivors might not yet have experienced symptoms because of infection late in the outbreak and therefore shorter follow-up interval. To reduce potential misclassification of exposure status, we restricted our analyses to Bundibugyo Ebola virus survivors classified as probable or confirmed and their seronegative contacts. Furthermore, the two sensitivity analyses assessing different exposure classifications did not substantially change our conclusions. Because serological testing was done more than 2 years after the outbreak, Bundibugyo Ebola virus antibodies might have waned, potentially resulting in misclassification of exposure status for some contacts. This potential misclassification would probably reduce our ability to detect significant differences. We were unable to locate ten Bundibugyo Ebola virus survivors, and two had relocated. The loss to follow-up could have been related to their health status; the survivors could have had worse health and relocated to receive better care, or alternatively could have had better health and were therefore more mobile. However, we expect the effect on the results to be negligible because only 6% of the survivors were lost to follow-up.

Our findings support those from various studies of post-infectious sequelae, especially arising from severe disease. Investigation into the mitigation of chronic complications is needed for Ebola virus infection and many other infectious diseases. Our study included only adult survivors; because the long-term health effects experienced by children after severe disease are probably different from those of adults, additional studies are needed to establish the health needs for children who survive Ebola virus disease. Continued research efforts are needed to yield benefit for survivors of the continuing Ebola virus disease epidemic in west Africa as well as survivors of previous outbreaks.

Contributors

DVC, HK, MM, MAE, ANH, MH, LH, and MR designed the clinical study. HK, MM, SW, LL, NLM, and MLR implemented the study, including data collection and interpretation of the results. AT, MAE, LAE, AH, GGOJr, RJS, and LEH designed and did the laboratory testing. DVC analysed the data. All authors contributed to writing the report.

Declaration of interests

ANH, GGOJr, RJS, and LH received Department of Defence funding. All other authors declare no competing interests.

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