Student's Journal of Health Research Africa
e-ISSN: 2709-9997, p-ISSN: 3006-1059
Vol.7 No. 2 (2026): June 2026 Issue
https://doi.org/10.51168/sjhrafrica.v7i2.2342
Original Article

Understanding the epidemiology of Ebola Bundibugyo virus (EBV) in selected populations: a cross-sectional study in Buhendera and Bwamba counties, Bundibugyo district.

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Abstract

Background:

Bundibugyo district has an estimated population of 224,387 people. Ebola Bundibugyo was first identified in 2007, leaving over 150 people infected and over fifty-six deaths recorded, giving 40% fatality rate.

Methodology:

A cross-sectional study was carried out in the six study sites, selected from each county. Risk factors, distribution, endemicity, and mode of action and transmission of this strain were examined. Risk factors, distribution, and endemicity were assessed using systematic sampling, mode of action, and transmission, and the anatomy was examined using the WHO standards of Automated or semi-automated nucleic acid tests (NAT) and antibody-capture enzyme-linked immunosorbent assay (ELISA).

Results:

This research examines the general epidemiology of Ebola Bundibugyo among selected populations in Bughendera and Bwamba counties in Bundibugyo district. Bundibugyo had high cases, although these were not from the place where the first case was reported. The virus spread through movements, followed by Kasitu, who is believed to have reported the first case, Harugale, Busaru, Bubukwanga, Nduguto, and Karugutu. The majority of males (58.6%) said that they were involved in activities like collecting fruits from the forests, including hunting 41.4% were female who remained home waiting to cook what their male counterparts had brought from the forest. 72.2% reported that their relatives did not have access to the medical facilities. This indicates that healthcare facility accessibility was significantly related to EBV.

Conclusion:

The Ebola-Bundibugyo strain had a high fatality rate at 40%. The study revealed that cultural risk factors were more responsible for this distribution and severity of EBV.

Recommendations:

Ebola preventive messages should be incorporated into the routine health education scripts of the Ministry of Health of Uganda and the East African countries.

***Keywords**: Ebola, hemorrhagic fever, Bundibugyo, outbreak, laboratory-confirmed.*
***Submitted:** December 31, 2025 **Accepted:** April 29, 2026 **Published:** June 11, 2026*

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Introduction

What once started as superficial turned out to be the deadliest disease. The source of the first person who showed symptoms has remained unknown; however, according to the locals, a place called Kikyo can never be forgotten. One of the locals said that one person was hacked to death for being suspected of having brought spiritual powers into the family, which was killing the family members. What worsened the situation was the fact that whoever killed another on ground of him/her having brought witchcraft into the family ended up contracting the disease and developing the same signs and symptoms. The locals said that reports about they termed as a strange disease started in 2006. It should be remembered that this was a period when Uganda was about to host the CHOGM meeting. Sources at the district who were contacted say that the Ministry of Health declined to declare the outbreak as the deadly Ebola until the end of the meeting (Kaner & Schaack, 2016). The first case of Ebola hemorrhagic fever (EHF) outbreak caused by Bundibugyo Ebola virus was confirmed in Bundibugyo District, Uganda, in 2007. A doctor who carried out operations for intestinal perforations, Doctor Monday recalls seventy-two cases, of which only fifty-six cases were laboratory confirmed. Although signs and symptoms were largely nonspecific and similar to those of EHF outbreaks caused by Zaire and Sudan Ebola viruses, the proportion of deaths among those infected was lower at 40.02% (Francesconi et al., 2003)

The Ebola virus causes an acute, serious, and fatal illness. The virus family Filoviridae has three genera: Cuevavirus, Marburgvirus, and Ebolavirus. Within the genus Ebolavirus, five strains have been identified: Zaire, Bundibugyo, Sudan, Reston, and Taï Forest. The first three, Bundibugyo ebolavirus, Zaire ebolavirus, and Sudan ebolavirus, have been associated with large outbreaks in Africa. A subset of surviving persons who had a history of illness consistent with EHF but no acute-phase blood samples available for testing, a convalescent-phase blood sample was collected for laboratory confirmation by IgG ELISA (Yang et al., 2000). Laboratory testing was performed at the Uganda Viral Research Institute (UVRI) in Entebbe, and subsequent testing was performed on some samples at CDC, Atlanta (Kaner & Schaack, 2016). Most studies conducted so far have relied on the fifty-six confirmed cases and were conducted during the 2006/2007 outbreak (Yang et al., 2000). No known studies have been conducted after the outbreak. This research looks at the general epidemiology of this virus, with thirteen years of no further outbreaks.

Materials and methodology

Study design

This research was a cross-sectional study in which an observational design was used, where data were collected from the affected population at a single point. Exposure risk factors and outcomes were measured concurrently to identify correlations without manipulating any variables.

Description of study area

Geographical location of the study area

Bundibugyo District is located in Western Uganda. It comprised of Three Counties, Bughendera, Bwamba, and Ntoroko. However, in 2010, the government, through an act of parliament, carved Ntoroko County into a fully- fledged district. The district is located in the Western part of Uganda. It is bordered by Kabale district in the North-East, Kabarole district in the East and South-East, and to the West by the Democratic Republic of Congo. The district covers a total area of 2338 $km^{2;}$ of this area, 570 km^2 is covered by open water, swamps, and rivers, while 1243 km^2 is covered by mountains, forests, national parks (Semuliki and Rwenzori National Parks), and the forest reserves, 145 km^2 is covered by the game reserves, and 380 km^2 is used for agriculture. Only a small portion of land is left for agriculture. This area is mountainous (District Development Plan, 2006/2007- 2008/2009).

Geographical Information System (GIS)

Coordinates were taken from strategic points in each of the study areas, and overlays were carried out using a Google-generated site map. Data generated from the field were analysed using ArcGIS and a case assessment.
The map was drawn.

Statistical analysis

In order to understand the period of the first outbreak, we interviewed farmers where the first case is believed to have been identified. To be more accurate, a data viewing tool from NOAA was used to analyse the climatic risk factors. The interface enabled the researcher to access data in terms of temperature and rainfall by retrieving them retrospectively from NOAA (National Oceanic and Atmospheric Administration)

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Figure 1: Case analysis map of Ebola Bundibugyo (EBV)

(Google generated map using primary data and National Oceanic and Atmospheric Administration, 2018)

The most affected areas in the district were Kasitu, Busaru, Harugale, Nduguto, Bubukwanga, Bundibugyo town council, and Karugutu. Every point on the map represents an area mentioned above, with the number of confirmed cases, survival cases, and deaths **(Figure 1)**

Participatory Research Appraisal (PRA) Methods

Under this method, several tools were used, which included 7 interview guides, one in every parish, a questionnaire, and a review of the available data at the main hospital and the district department of the District Health Officer (DHO). Three types of questionnaires were used: one for the households that met the inclusion criteria, another for the local authorities, and another for the NGOs/CBOs like MSF, CDC, and COHASPA, which participated during the outbreak. Expert knowledge was taught by doctors who participated in handling patients, like Dr. Monday, who handled the first cases at a health Centre IV in Kikyo. Indigenous knowledge about the known reservoirs was obtained from the traditional leaders, who were commonly known as herbalists. The households were sampled used snow ball sampling because those that lost relatives had been identified by the time of this research. Cultural risk factors like sleeping near the dead by a relative, partner sleeping with the dead, washing the dead, preserving body parts, and performing cultural rituals were assessed to ascertain their contribution towards EBV. A questionnaire was used to gather data about these factors.

Mode of action and anatomical structure, as well as the biochemistry of Ebola Bundibugyo, were analysed using the IgG ELISA method on preserved samples at UVRI, as well as reviewing literature from CDC publications and the international procedure of studying the structures of viruses.

Table 1: Strain Information about the Ebola Bundibugyo virus

l	**Bundibugyo virus/H.sapiens-tc/UGA/2007/Butalya-811250**
Organism:	Bundibugyo ebolavirus
Taxonomy:	Filoviridae -> Ebolavirus -> Bundibugyo ebolavirus
GenBank Host:	Homo sapiens

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Host:	Human
Isolation Country:	Uganda
Collection Date:	2007-11
Comments:	Bundibugyo ebolavirus isolate Bundibugyo virus/H. sapiens-tc/UGA/2007/Butalya-811250, complete genome.

(Niikura *et al*., 2001)

Strain type of Bundibugyo nature. Samples collected confirmed a different strain. Naming was done according to the place of the outbreak. This strain became different from Zaire, but with high fatality **(Table 1)**

Table 2: Genome: NC_014373

GenBank Definition:	**Bundibugyo ebolavirus isolate Bundibugyo virus/H. sapiens-tc/UGA/2007/Butalya-811250, complete genome.**
GenBank Sequence Accession:	NC_014373
Sequence Length:	18940
Sequence Status:	Complete
Sequence:	View Nucleotide Sequence and design PCR primers
Number of Proteins:	9
Organism Name:	Bundibugyo ebolavirus
Isolation Source:	blood
Mol Type:	viral cRNA
GenBank Host:	Homo sapiens
Host:	Human
Isolation Country:	Uganda
Collection Date:	2007-11

(Cao, Bai, Wang, & Che, 2018)
The distinct features of the Ebola Bundibugyo Virus Sequence length, number of proteins, the Mol type, among others, clearly outline the genome composition **(Table 2)**

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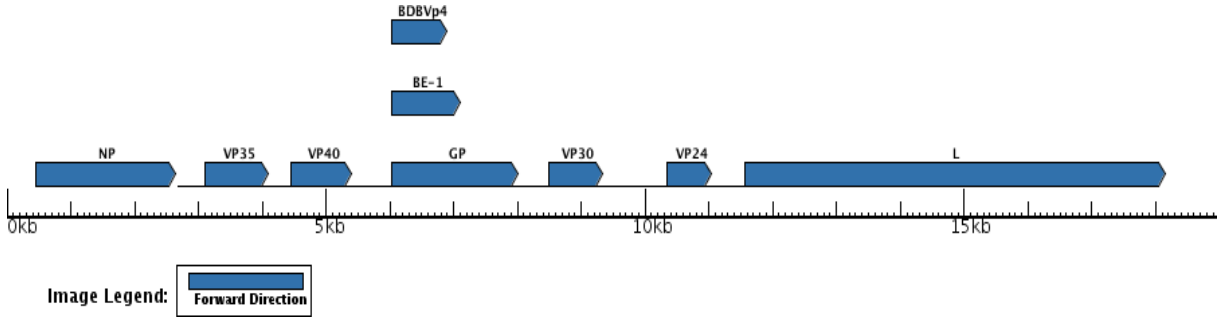
Figure 2: Genome Image Map of Ebola Virus



(Niikura et al., 2001)

Protein alignment NP, VP35, VP40, GP, VP30, BDBVp4, BE-1, and VP24, among others, gives the virus its actionality properties. The structure of GP2 by X-ray crystallography revealed that GP2 contains a central triple-stranded coiled coil followed by a disulfide-bonded loop (Figure 2). GP2 could bridge two membranes to initiate membrane fusion (46). The trimeric crystal structure of surface glycoprotein (GP) is also required to initiate attachment and fusion of viral and host membranes that further bind to a neutralizing antibody, KZ52, in humans (42). Human lysosomal cholesterol transporter Niemann-Pick C1 (NPC1) fulfills a cardinal property of viral receptors and binds specifically to viral GP **(Figure 2).**

Table 3: Protein Information of Ebola Bundibugyo Virus

Gene Symbol	Protein Product Name	ViPR Locus ID	CDS Start	CDS End	NCBI Gene ID	Locus Name	Source
NP	nucleoprotein	NP	458	2677	9487269	BDBVp1	GenBank
Gene Symbol	Protein Product Name	ViPR Locus ID	CDS Start	CDS End	NCBI Gene ID	Locus Name	Source
VP35	polymerase complex protein	VP35	3108	4133	9487263	BDBVp2	GenBank
VP40	matrix protein	VP40	4461	5441	9487264	BDBVp3	GenBank
GP	second secreted glycoprotein	BDBVp4	6021	6930	9487265	BDBVp4	GenBank
GP	small secreted glycoprotein	BE-1	6021	7142	9487265	BDBVp4	GenBank
GP	spike glycoprotein	GP	6021	8050	9487265	BDBVp4	GenBank
VP30	minor nucleoprotein	VP30	8496	9365	9487266	BDBVp5	GenBank
VP24	membrane-associated protein	VP24	10335	11090	9487267	BDBVp6	GenBank
L	RNA-dependent RNA polymerase	L	11567	18199	9487268	BDBVp7	GenBank

(Moghadam, Omidi, Bayrami, Moghadam, & SeyedAlinaghi, 2015) 9 protein types with unique characteristics further distinguish the Ebola Bundibugyo Virus from other strains **(Tables 2 and 3)**

Review of Secondary Data

The study reviewed literature about the proportion of deaths and clinical features in Bundibugyo Ebola virus infection, Uganda, and the available national guides on Ebola Virus, as well as the documented statistics in the archive of the office of the district health officer.

The study reviewed literature about the progress already made by different researchers, such as CDC, MSF, WHO,

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and other reports available in the office of the DHO. Seventeen publications were reviewed, five of which were specifically on Ebola Bundibugyo. Others were the international guidelines on the testing and analysis of the Ebola virus, as set by the CDC and WHO, and the shipping regulations of samples.

Ethical consideration

A letter of recommendation was issued by the Department of Biological Sciences in June 2023 to the Kyambogo University Research and Ethics Committee. An approval letter was issued in February 2024.

RESULTS

The research used quantitative methods of data collection. Mapping of the areas which were affected by the outbreak was done by capturing, generating, and analysing using ArcGIS. A case Analysis Map (Fig 1) was generated using coordinates taken from strategic points in each of the study areas, and overlays were carried out using a Google-generated site map.

Table 4: EBOLA BUNDIBUGYO VIRUS INFECTION PER SUBCOUNTY AS OF 2007

S.no	SUB-COUNTY	CONFIRMED CASES	SURVIVED	DIED
1	KASITU	10	4	2
2	BUSARU	6	5	1
	HARUGALE	7	4	1
4	NDUGUTO	5	2	3
5	BUBUKWANGA	6	4	2
6	BUNDIBUGYO TC	17	4	6
7	KARUGUTU	5	3	2
	Total	56	26	17

The above table represents cases per subcounty. Bundibugyo had high cases, although it was not the place where the first case was reported. The virus spread through movements, followed by Kasitu, who is believed to have reported the first case, Harugale, Busaru, Bubukwanga, Nduguto, and Karugutu **(Table 4)**

Risk factors

Several risk factors were investigated using Participatory Research Appraisal (PRA) tools such as the questionnaires, interview guides, observation check lists, and key informant guides, among others.

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Influence of gender on EBV

The study examined the impact of gender within households on EBV as one of the socio-demographic risk factors. This was done through cross-tabulation analysis.

The majority of males (58.6%) said that they were involved in activities like collecting fruits from the forests, including hunting 41.4% were female, who remained home waiting to cook what their male counterparts had brought from the forest

Table 5: Influence of gender on EBV

Variable		Frequency	Percentage
A. Influence of gender on EBV			
i.	Male	225	58.6
ii.	Female	159	41.4
Total		**384**	**100**

In this table, Males were more influential in all aspects, such as distribution, because of the cultural superiority of males in this society. Males went hunting, and they made sure that cultural norms were observed **(Table 5)**

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However, to conclude on this, a chi-square test statistic was run.

The chi-square tests (x^2=4.520 and p≤1") revealed a significant influence of the gender of the household on the EBV, given that the P-Value 0.034 was less than the customary 0.05 at 95% level of significance. This led to rejection of the null hypothesis, and the conclusion was made that household gender had a significant impact on EBV.

Table 6: Chi-Square Tests on the Influence of gender on Ebola Bund

	Value	df	Asymp. Sig. (2 sided)	Exact sig. (2-sided)	Exact sig (1-sided)
Pearson Chi-square	4.52	1	0.33		
continuity correction	4.088	1	0.043		
likelihood ratio	4.535		0.33		
Fisher's extract test					
Linear -by-linear					
Association	4.508	1	0.34	0.037	0.21
N valid cases	380				
a. Computed for a 2x2 table					
b. 0 cells (0.0%) have expected more than 5. The minimum expected counts is 75.19					

Household gender was influential in the risks of Ebola. More males meant manpower for hunting and gathering wild fruits, thus more susceptibility **(Table 6)**

Accessibility to the health centers

The influence of healthcare service accessibility on EBV was tested using cross-tabulation statistics analysis, as summarized in the table below. 72.2% reported that their relatives did not have access to the medical facilities. This indicates that healthcare facility accessibility was significantly related to EBV.

Table 7: Accessibility to the health centers

VARIABLE	FREQUENCY (f)	TOTAL	X^2	p-value
A.	**Yes**	**No**	11.039	1
Accessibility				
Yes	135	161 296		
	(86.5%)	(72.2%) (78.1%)		
No	21	62 83		
	(13.5%)	(27.8%) (21.9%)		
Total	**156**	**223 379**		
	100%	**100% 100%**		

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Based on the fact that specific isolation centers were erected at Kikyo, Nyahuka, and Bundibugyo Hospital, these were not enough to handle the cases. Some suspected cases were afraid of long stretches to the isolation centers. Several cases were not fully taken into account since they did not access the isolation centers **(Table 7)**

This was further investigated using chi-square test statistics. According to the chi-square test statistics (x^2=8.992) that was carried out, the observed significance level was 0.001, which is less than the customary 0.05. Based on the fact that the "P≤0.001" is less than 0.05, the null hypothesis is rejected and a conclusion drawn to the effect that accessibility to healthcare facilities had a significant positive influence on EBV.

Table 8: Chi-Square Tests on the Influence of Access to Health Services on Ebola Bund

Table 8: Chi-Square Tests on the Influence of Access to Health Services on Ebola Bund	Value	df	Asymp. Sig. (2-sided)	Exact sig. (2-sided)	Exact sig (1-sided)
Pearson Chi-square	11.037b	1	0.001		
continuity correction	10.216	1	0.001		
likelihood ratio	11.549	1	0.001		
Fisher's extract test					
Linear -by-linear					
Association	11.008	1	0.001	0.001	0.001
N valid cases	379				
a. Computed for a 2x2 table					
b. 0 cells (0.0%) have expected more than 5. The minimum expected counts is 34.16					

A confirmation that access to health services influenced the general epidemiology of the Ebola Bundibugyo Virus. Even where access to health services was possible, the availability of health professionals to patient ratio, as well as the readiness of the health workers, remained a big challenge **(Table 8)**

Impact of Religion on EBV

Household religious affiliation was one of the personal risk factors taken into account in understanding the risk factors for EBV. This was achieved by running a cross-tabulation between religious affiliation and prevalence of cases of EB. This revealed a significant influence of religion on the EBV, where Catholics were found to experience more cases, followed by SDA (24.5%), Islam (23.8%), among others. This was, however, proved further by running chi-square test statistics. At chi-value (x^2 = 24.918), the observed significant level (P- Value) was "p≤0.000", less than the expected 0.05 at 95% confidence level. Based on this figure, the null hypothesis was rejected and a conclusion drawn to the effect that religious affiliation had a significant influence on EBV.

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Table 9: Impact of religion on EBV

What is your religious affiliation?		Do you have a person suffering/dead from Ebola?				Total	
		Yes		No			
	Islam	26	17.7%	14	6.5%	40	34.8%
	Catholic	36	24.5%	32	14.9%	68	18.8%
	Anglican	31	21.1%	38	17.7%	69	19.1%
				90			
	Pentecostal	35	23.8%	41.9%		34	34.5%
				41			
	SDA	19	12.9%	19.1%		60	16.6%
				215			
Total		147	100%	100%		362	100%

Religion presented a highly significant influence on Ebola Bundibugyo, the number of respondents who answered yes to having had an Ebola patient in each religious affiliation, Islam at 17.7%, Catholic at 24.5%, Anglican 21.1%, Pentecostals at 23.8%, with SDA showing exceptional at 12.7% **(Table 9)**

Impact of occupation

The results of the regression coefficients analysis revealed a sample slope of 2.453 standard error units, 0.014 below the hypothesized value of 0. In view of the fact that this is a most unlikely experience where the observed significance value is less than the expected 0.05 at 95% confidence interval, the null hypothesis is rejected. It becomes visible that household occupation had a significant positive impact on EVB. It was revealed that the first outbreak was reported from a farmer in a place called Kikyo.

Table 10: Regression coefficients on the impact of occupation on Ebola Bund. Virus

	Unstandardized Coefficients		Standardized Coefficients		T	Sig.	95% Confidence Interval for B	
Model		B	Std. Erro r	Beta			Lower Bound	Upper Bound
1	(Constant)	1.529	.052		29.549	.000	1.427	1.630
	What is your occupation?	3.517E-02	.014	.138	2.453	.015	.007	.063

A Dependent Variable: Did you have someone suffering from EBV?

In this table, it is revealed that farmers were blamed for the outbreak; the farmers in Kikyo were closer to the reservoirs. Maize is a palatable food for the monkeys; it is believed that the first case broke out from a maize farmer **(Table 10)**

Impact of Income Earned on EBV

The regression coefficients statistics were run. From the table, the observed significance level 0.000 was less than the expected significance level of 0.05 at 95% confidence interval. In view of the fact that this is a most improbable experience, the null hypothesis that income had no significant impact on EBV is rejected. Therefore, considering this result, the researcher strongly concluded that household income had a positive impact on the EBV. It was revealed that EBV started in a low-income household.

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Table 11: Regression coefficients analysis between income and EBV occurrence

Model		Unstandardized Coefficients		Standardize Coefficients	dT	Sig.	95% Confidence Interval for B	
		B	Std. Error	Beta			Lower Bound	Upper Bound
1	(Constant)	2.036	.065		31.283	.000	1.908	2.164
	What is your average monthly income in Uganda shilling?	-.172	.024	-.356	-7.249	.000	-.218	-.125

a Dependent Variable: Did you have someone suffering from EBV?

From the table, income status determined the hunting urge. Low income meant more preferences for hunting than medium income status. Peasants hunted more frequently than other occupations. EBV started from a low- income household **(Table 11)**

Impact of family size

Respondents were allowed to state the number of people in their family in ranges and if they were affected by EBV. In the regression coefficient, the observed significance level (0.002) is less than the customary significance level (0.05) at 95% confidence interval. Given this outcome, the null hypothesis was rejected and a conclusion drawn to the effect that there was a linear relationship between household size and EBV.

Table 12: Regression coefficients on the influence of household size on EBV

Model		Unstandardized Coefficients		Standardized Coefficients	T	Sig.	95% Confidence Interval for B	
		B	Std. Error	Beta			Lower Bound	Upper Bound
1	(Constant)	1.901	.099		19.125	.000	1.706	2.097
	How many people live in this house?	-.161	.052	-.160	-3.099	.002	-.262	-.059

A Dependent Variable: Did you have someone suffering from EBV?

In the table, households with large numbers were more susceptible to EBV than households with small family members **(Table 12).**

Influence of the tribe

The cross tabulation indicates inconsistent results where the majority of Bakonzo (37.6%) reported experiencing more cases, and at the same time, they were the largest tribal group who reported not experiencing low cases (34.1%). This means that the tribe had no significant influence on EBV. This gave room for further investigation through running chi-square test statistics in the table below. The observed significance level, at chi-value 36.559, "P≤0.625" greater than the expected 0.05. Based on this result, the null hypothesis is accepted, and the conclusion is made that tribes or tribal differences had no significant influence on EBV

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Table 13: Chi-Square Tests on the Influence of tribe on Ebola Bund

Table 13: Chi-Square Tests on the Influence of tribe on Ebola Bund	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-square	36.559a	5	0
continuity correction	38.745	5	0
likelihood ratio			
Fisher's extract test			
Linear -by-linear			
Association	0.239	1	0.625
N valid cases	380		
a. Computed for a 2x2 table			
b. 0 cells (0.0%) have expected more than 5. The minimum expected counts are 8.68			

Tribe did not have a significant impact; however, it depended on the proximity to the inhabitants of the reservoirs. (Table 13) Education level.

The level of education of the households was taken into consideration. This was proved by running a cross-tabulation statistic, and the results of the cross-tabulation revealed a contradiction where the majority of households with tertiary education (55.9%) reported cases, and at the same time, they were the greatest percentage reported not having experienced any case. This indicates that the education level was not significant. However, before concluding the above cross-tabulation results, a chi-square test statistic (x^2=6.576) was run, which revealed that the "P≤0.058" was greater than the customary 0.05. This led to the acceptance of the null hypothesis that household education level had no significant influence on EBV

Table 14: Cross-tabulation of cultural risks

What are your cultural practices?		Do you have a person suffering/dead from Ebola?				Total	
		Yes		No			
	sleep near the dead	27	17.2%	104	47.5%	131	34.8%
	partner sleeps with the dead	90	57.3%	62	28.3%	152	40.4%
	Washing the dead	16	10.2%	20	9.1%	36	9.6%
	Preserving of body parts	11	7.0%	23	10.5%	34	9.0%
	Performing burial rites	13	8.3%	10	4.6%	23	6.1%
Total		157	100%	219	100%	376	100%

In this table, lying with the dead as a cultural practice was responsible for the distribution of EBV. The situation was worsened before a public declaration of the outbreak. Cultural practices played a big role in the general epidemiology of EBV **(Table 14)** Of the cultural risk factors, a partner sleeping where his/her loved one who has been laid in the house took a center stage at 40.4%, and still a relative sleeping near the dead became the second at 34.8%, and then washing the dead at 9.6%

Climatic risk factors

Being the first known outbreak in the Bundibugyo district, there was no retrospective reference, and the researchers carried out an exploratory investigation of the climatic risk factors.

The district experiences bimodal rainfall patterns. The first rains are short and occur during March-May, and a longer period from August-November. Annual rainfall ranges are less than 800mm and is great influenced by altitudes.

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e-ISSN: 2709-9997, p-ISSN: 3006-1059
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Rainfall distribution and reliability enable agricultural activities to take place throughout the year, mostly at the foot and slopes of Mount Rwenzori.

Table 15: Rainfall distribution for Bundibugyo district.

Month	Jan	Feb	Mar	Apr	May	June	July	Aug	Sept	Oct	Nov	Dec
Depth/mm	40	60	140	210	140	80	60	130	170	220	160	90

National Oceanic and Atmospheric Administration, 2017. The district has two harvesting seasons, in May and July/August. The first laboratory confirmed case was announced in June, at the peak of harvesting. This is the time when interaction/competition for the farm produces between the reservoirs, such as Monkeys and fruit bats, and farmers is high **(Table 15)**

Temperature

There is a wide temperature variation influenced by altitude, fluctuating from very high in the plains to zero degrees in the higher Mountains. The temperature in this area is influenced by relief. Mean annual maximum temperature varies between 27.5 and 40.0 degrees Celsius in Jan and February, while the average annual minimum temperature varies between 12.5 and 16.0 degrees Celsius in September to November. Based on the findings, it was revealed that the out-break tool place during the planting season. The above conditions were favorable for the wild animals like Monkeys come out and pick some of the planted seeds. In such cases, they interact with the communities as revealed by one of the informants we talked to.

For purposes of accuracy, a link between forest distraction and the general epidemiology of the Ebola outbreak in Bundibugyo was examined. An Ebola map showing the link between forest destruction and the outbreak can be seen using the figure below (you will need Google Earth Pro software to view this map)

Figure 3: Google map, forest destruction, and the outbreak of Ebola Virus in selected populations in Africa

(Mutebi *et al.*, 2012)

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Vol.7 No. 2 (2026): June 2026 Issue
https://doi.org/10.51168/sjhrafrica.v7i2.2342
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Like many forested areas in the Democratic Republic of the Congo, Mount Rwenzori has suffered great destruction, leaving only 40% remaining area with forest cover. This map was generated by comparing forest cover destruction with the frequency of Ebola outbreaks. Mount Rwenzori stretches from Kasese district to Fort-Portal, Bundibugyo, up to the far East of DRC. Several Ebola outbreaks have been recorded along this stretch, including the 2007 Bundibugyo outbreak. Forest cover destruction has proven to create great exposure to Ebola outbreaks. This is due to the fact that wild animals come closer to the communities **(Figure 3).**

In many outbreaks in Africa, cultural risk factors remain the main means by which the virus is passed from one person to another. For partners, one partner died of the virus, culture recommended that his/her partner sleep on the same beddings as the dead; furthermore, for those who were not married, a relative would sleep near the dead. Washing the dead before burial was another cultural risk factor responsible for the distribution of EBV. CDC researchers identified cultural risk factors as the major risk to the outbreak of Ebola in West Africa.

Distribution of EBV

Table 16: Ebola Bundibugyo virus infection per subcounty as of 2007

S.no	SUB-COUNTY	CONFIRMED CASES	SURVIVED	DIED
1	KASITU	10	4	2
2	BUSARU	6	5	1
	HARUGALE	7	4	1
4	NDUGUTO	5	2	3
5	BUBUKWANGA	6	4	2
6	BUNDIBUGYO TC	17	4	6
7	KARUGUTU	5	3	2
	Total	56	26	17

Kasitu, despite being the area where the first case was confirmed, the distribution was aided by the movement of people. Bundibugyo, being the central place, received many people from different places. This was confirmed by more cases than in other places **(Table 16)**

Student's Journal of Health Research Africa
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Vol.7 No. 2 (2026): June 2026 Issue
https://doi.org/10.51168/sjhrafrica.v7i2.2342
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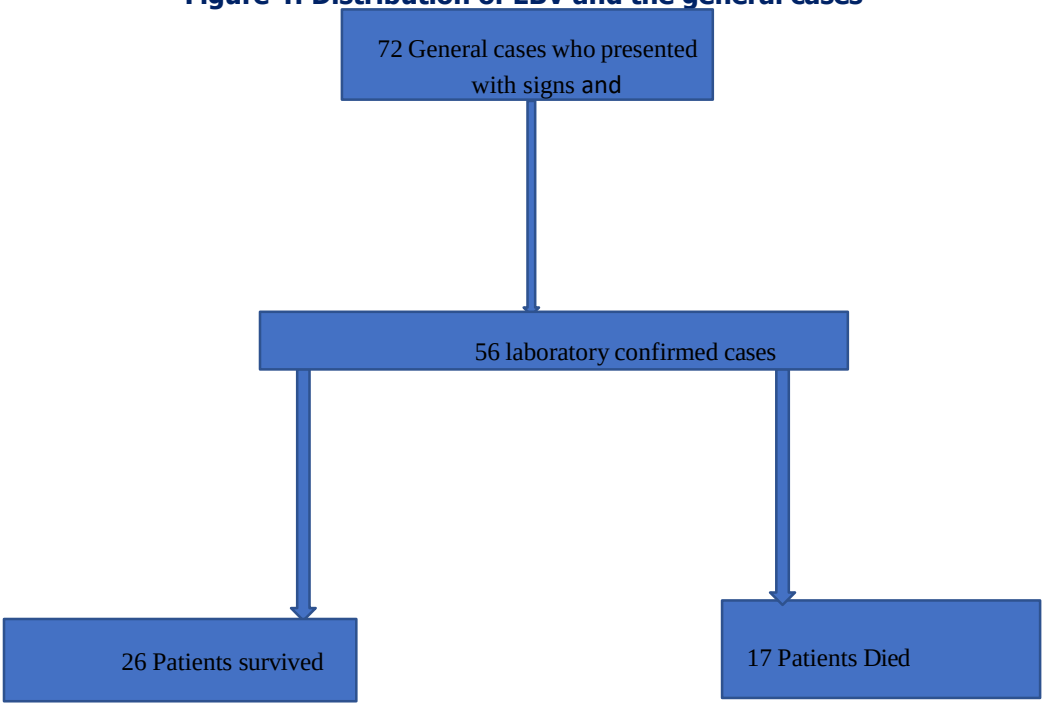
Figure 4: Distribution of EBV and the general cases



According to the current details from the locals, 105 cases were recorded. Officially, 72 cases were reported at the district level; clinically, 56 were confirmed cases. 26 patients survived, and 17 died **(Figure 4).** *Residents say that many cases were not recorded (Key Informant, 2008)***Distribution** was dependent on the proximity to the hotspot areas like Kiryo, Ngamba, and Ntandi. Cases in Bundibugyo town council were mainly acquired through contact with the infected persons. Late Kule Joshua, a senior clinical officer, as well as Dr. Kule Jonah, a senior nurse, and Bulimpikya Rose are believed to have contracted the virus from the patient he attended to. Others, like late Maate Asanasio (Orthopedic Clinical Officer), Jeremiah Muhindo, who is believed to have infected late Masereka Enock and Muhindo Jovia, Kiiza Johnson (Enrolled Nurse), among others, are believed to have attended the burials of the dead and attended to their sick relatives. Bundibugyo Town Council recorded more cases, followed by Kasitu. Cultural risk factors were the primary predisposing factors to the distribution of EBV.

Mode of Action

Like the rest of the strains, EBV's mode of action was from the reservoir to the person, then from the infected person, not another. Any contact with the fluids of the person who has presented signs and symptoms puts a person at high risk of contracting the virus. The entry points into the body were the vagina, vagina penis, mouth, ears, eyes, nose, cuts, among others. When it enters the body, EBV synthesises a virion glycoprotein (GP) which induces cytotoxic effects in human endothelial cells. This effect is mapped to a serine–threonine-rich, mucin-like domain of type I transmembrane glycoprotein, one of seven gene products of the virus. The genome transfer of GP into human or porcine blood vessels then causes massive endothelial cell loss within 48 hours, which leads to a substantial increase in vascular permeability. Deletion of the mucin-like region of GP removed the effects without affecting protein function. This indicated that GP, through its mucin domain, was the viral determinant of EBV pathogenicity or mode of action inside the host and contributed to hemorrhagenicity during the infection.

Student's Journal of Health Research Africa
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Vol.7 No. 2 (2026): June 2026 Issue
https://doi.org/10.51168/sjhrafrica.v7i2.2342
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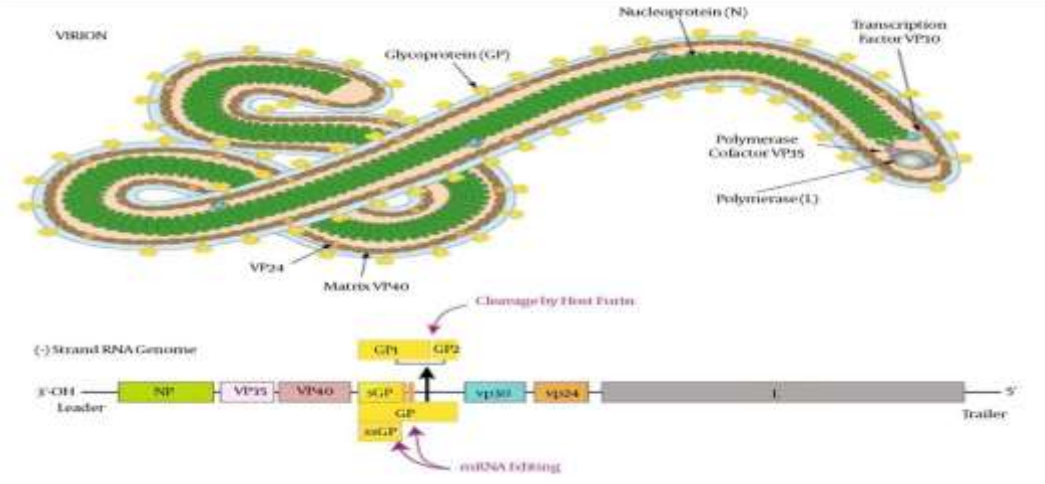
Table 17: Anatomical features of EBV that give its mode of action

Recombinant	Subtype	Anatomical features
BDBV	Bundibugyo, strain Uganda 2007	Small Glycoprotein (sGP) Amino acids 33-325 produced in mammalian HEK293 cells incorporates a C-terminal 6xHis tag Envelope glycoprotein is initially produced as a precursor known as pre-GP, cleaved by furin into two subunits, GP_1 and GP_2, which remain associated through a disulfide linkage between Cys53 of GP_1 and Cys609 of GP_2 The heterodimer assembles into a 450-kDa trimer at the surface of nascent virions. Action by attachment, fusion, and entry of target cells (GP1) is derived from the GP1 sequence, ACI28624, expressing Ile33 – Gln304 (GP1) is fused with a polyhistidine tag at the C-terminus, and has a calculated MW of 31.8 kDa protein in HEK293 cells DTT-reduced Protein migrates as 40-60 kDa

CDC/VRI,2018 Preserved specimen Complex, only structurally studied under an electron microscope. GP proteins give their mode of action **(Table 17)**

Figure 5: Anatomical structure of EBV as seen under an electron microscope



(Holmes, Dudas, Rambaut, & Andersen, 2016)

Student's Journal of Health Research Africa
e-ISSN: 2709-9997, p-ISSN: 3006-1059
Vol.7 No. 2 (2026): June 2026 Issue
https://doi.org/10.51168/sjhrafrica.v7i2.2342
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Anatomically helical, single-stranded, unsegmented RNA negative sense, Envelope glycoprotein is initially produced as a precursor known as pre-GP, cleaved by furin into two subunits, GP1 and GP2, which remain associated through a disulfide linkage between Cys53 of GP1 and Cys609 of GP2 **(Figure 5)**

Discussion

Generalizability

In general, almost all assessed risk factors had a prevalence impact on the epidemiology of the Ebola Bundibugyo virus. The risk factors assessed were gender, occupation, income level, tribal education level, medical, religious, and climatic. Findings revealed a significant impact of gender (Male: female) of the household on the EBV (Moghadam et al., 2015). Furthermore, there was a significant influence of religion on the EBV, where Catholics were found to experience more cases, followed by SDA (24.5%), Islam (23.8%), among others (Kaner & Schaack, 2016). Household occupation had a significant positive impact on EVB. It was revealed that the first outbreak was reported in a farmer in a place called Kikyo (MacNeil *et al.*, 2010). Household income had a positive impact on the EBV. It was revealed that EBV started from a low-income household (Formenty, 2014). There was a linear relationship between household size and EBV (Moghadam et al., 2015). 72.2% reported that their relatives did not have access to the medical facilities. This indicates that healthcare facility accessibility was significantly related to EBV (Dietz, Jambai, Paweska, Yoti, & Ksiazek, 2015). Tribal factors were investigated, where the majority of Bakonzo (37.6%) reported experiencing more cases, and at the same time, they were the largest tribal group who reported not experiencing low cases (34.1%) (Moghadam et al., 2015). However, education level had no significant effect (Francesconi *et al.*, 2003). On climatic risk factors, it was revealed that a wide temperature variation influenced by altitudes fluctuates from very high at the plains to zero degrees in the higher Mountains. The temperature in this area is influenced by relief. Mean annual maximum temperature varies between 27.5 and 40.0 degrees Celsius in Jan and February, while the average annual minimum temperature varies between 12.5 and 16.0 degrees Celsius in September to November. Based on the findings, it was revealed that the outbreak occurred during the planting season. The above conditions were favorable for the wild animals, such as monkeys, to come out to pick out some of the planted seeds. In such cases, they interact with the communities as revealed by one of the informants we talked to (Camacho et al., 2014). *This disease was brought by a farmer who picked a dead monkey, took it home to eat, and his entire family died of the disease (Key Informant, 2017)*

Cultural risk factors remain the main means by which the virus is passed from one person to another. For partners, one partner died of the virus, culture recommended that his/her partner sleep on the same beddings with the dead; furthermore, for those who were not married, a relative would sleep near the dead. Washing the dead before burial was another cultural risk factor responsible for the distribution of EBV. CDC researchers identified cultural risk factors as the major risk to the outbreak of Ebola in West Africa (Selvaraj, Lee, Harrell, Ivanov, & Allegranzi, 2018). In fact, risk factors determined the trend of distribution. In areas where the virus was high, risk factors were more predisposing. To be specific, cultural risk factors were responsible for the secondary distribution of the virus (Formenty, 2014).

The mode of action was dependent on the genome of the virus. This virus is filamentous like other strains, under the filo family, ranging from 80 to 800-1000nm enveloped ssRNA, negative sense with a helical Genome. The mode of action was based on the fact that it replicated efficiently, producing large amounts of virus in monocytes, macrophages, dendritic cells, and other cells. Viral cytopathogenesis causes extensive tissue necrosis in parenchymal cells of the liver, spleen, lymph nodes, and lungs. The breakdown of endothelial cells leading to vascular injury and hemorrhage (Yang *et al.*, 2000).

Conclusions

1. The study revealed that cultural risk factors were more responsible for this distribution and severity of EBV. Cultural practices like sleeping with the dead, sleeping near the dead, preserving body parts, among others, posed a high risk to the communities. The period from June onwards, as revealed by NOAA, was suitable for the reservoirs to interact closely with the communities, especially the farmers and hunters. The research further revealed a significant connection between trade in wild meat and the general epidemiology of EBV. Surprisingly, family size had a significant influence on the epidemiology of EBV. Although there is still a need for more investigation as to why family size determines the epidemiology of EBV, the larger the family size, the faster the virus spreads through the entire family. Family size meant more need for food, and especially the delicacies. This was revealed to me by the Chi-square test. In general, the level of education and

Student's Journal of Health Research Africa
e-ISSN: 2709-9997, p-ISSN: 3006-1059
Vol.7 No. 2 (2026): June 2026 Issue
https://doi.org/10.51168/sjhrafrica.v7i2.2342
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tribe did not have any impact on the Epidemiology of EBV, while Family size, level of income, occupation, religion, access to medical facilities by the patients, and gender were revealed to have had a significant influence on the general epidemiology of EBV.

2. The study further revealed that of the seventy-two cases, Kasitu had the most cases, followed by Budibugyo Town Council and Harugale, respectively. Distribution also depended on the period a confirmed case was identified, and feedback as well as health education were conducted. The CHOGM meeting was also cited by one of the key informants as one of the reasons why the government delayed announcing the situation as an outbreak of the deadly Ebola virus.
3. The study revealed that EBV's mode of action inside the host was according to the protein molecule it contained and its ability to infect the endothelial cells. When it enters the body, EBV synthesises a virion glycoprotein (GP) which induces cytotoxic effects in human endothelial cells, mapped to a serine– threonine-rich, mucin-like domain of type I transmembrane glycoprotein, one of seven gene products of the virus. The genome transfer of GP into human or porcine blood vessels then causes massive endothelial cell loss within 48 hours, leading to a substantial increase in vascular permeability, and deletion of the mucin-like region of GP removed the effects without affecting protein function.
4. IgG ELISA method (Human Total Immunoglobulin G (Hu *IgG*) *ELISA* quantified the Hu *IgG* in human serum or plasma) on preserved samples revealed a small virion whose genome contained sGP incorporating a C-terminal 6xHis tag, enveloped with a MW of 31.8 kDa.

Limitations of the study

The findings of this research were limited to the two counties of Bughendera and Bwamba, in the six study sites, selected from each county; these were: Bubukwanga, Nyahuka Town Council, Bubandi, Mirambi, Kirumya, and Bundibugyo Town Council in Bwamba County and Harugale, Bukonzo, Ngamba, Sindila, Kasitu, and Ntandi in Bughendera County.

Limitations of the study

In the course of the research, several limitations were encountered, including the geographical terrain being mountainous, time constraints, and the inability of the respondents to respond to the research tool, especially the questionnaire and interview guides. However, these were handled by ensuring the use of the local research assistants from this area. The research objectives were explained to the respondents, and a high level of confidentiality was ensured.

Recommendations

Ebola preventive messages should be incorporated into the routine health education scripts of the Ministry of Health of Uganda and the East African countries.

Sources of funding

This research was funded by the researcher with help from the Cuban Doctors in Zanzibar, the State University of Zanzibar, the Kyambogo University Department of Biological Sciences, and friends.

Conflict of interest

There is no conflict of interest known. Where materials were used, the sources have been cited.

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Student's Journal of Health Research Africa
e-ISSN: 2709-9997, p-ISSN: 3006-1059
Vol.7 No. 2 (2026): June 2026 Issue
https://doi.org/10.51168/sjhrafrica.v7i2.2342
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University (Original). He is a researcher.

Acknowledgment

Special thanks go to the UVRI for allowing us to use their archives, MSF for training me in sample handling and case identification and isolation, CDC for allowing us to use their specialised laboratories and staff, and WHO for sponsoring this research. We are thankful to NOAA for providing the satellite maps and images dating back from 2018-2006. These helped in determining the climatic risk factors and the trend in climate change.

Running the samples at UVRI and at CDC was a rare and yet unique experience that not only imparted knowledge but also installed unique skills in me. I wholeheartedly thank you all. Like other threatening diseases, the Ebola virus has been contained through research and innovations, and sustainable preventive and control measures shall be developed.

Data Availability Statement

The data used to support the findings of this study are restricted by the Research and Ethics Committee at UVRI to protect PATIENT PRIVACY. Data are available from the Department of Biological Sciences of Kyambogo University, P.0 Box 1, Kyambogo, Kampala, Uganda, for researchers who meet the criteria for access to confidential data through Hindawi Publishing Journal of this article.

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Student's Journal of Health Research Africa
e-ISSN: 2709-9997, p-ISSN: 3006-1059
Vol.7 No. 2 (2026): June 2026 Issue
https://doi.org/10.51168/sjhrafrica.v7i2.2342
Original Article

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PUBLISHER DETAILS:

Student's Journal of Health Research (SJHR)
(ISSN 2709-9997) Online
(ISSN 3006-1059) Print
Category: Non-Governmental & Non-profit Organization
Email: studentsjournal2020@gmail.com
WhatsApp: +256 775 434 261
Location: Scholar's Summit Nakigalala, P. O. Box 701432, Entebbe Uganda, East Africa