Immunoglobulin G Seroprevalence of Lassa Fever Virus Among Individuals Residing Along the Banks of River Niger in Anambra State, Nigeria

Umezurike Kingsley Chimuanya[1], Chukwuma Oluchukwu Mariagoretti[1,2], Ochiabuto Mary Barbara[1], Manafa Patrick Onochie[1], Ntum Ifeanyichukwu Michael[1], Chukwuma George Okechukwu[1,2]

Abstract

Background – Lassa fever (LF) is an acute viral haemorrhagic fever caused by the Lassa virus belonging to the Arenaviridae family. It is a zoonotic infection that is widespread in West Africa. Transmission occurs through exposure to rodent excreta and secretions and via the bodily fluids of an infected person. This study aimed to determine the Lassa fever-specific IgG seroprevalence among individuals who reside along the banks of River Niger in Anambra state, Nigeria.

Materials and methods – Ethical approval was obtained from the ethics committee of the College of Health Sciences, Nnamdi Azikiwe University Awka. Informed consent was sought and a questionnaire was administered. For this study, 90 subjects were recruited from the Okpoko community of Ogbaru LGA of Anambra state, Nigeria. Blood samples were collected and analysed at the Molecular Research laboratory using the Enzyme-linked Immunosorbent Assay (ELISA) technique to screen for LF-specific IgG.

Results – LF-specific IgG was detected in 14 out of 90 Subjects, giving a prevalence rate of 15.6%. In addition, some risk factors were ascertained, and the relationship between LF IgG seroprevalence and age, gender, and Body Mass Index (BMI) were assessed though there were no significant relationships.

Conclusion – A significant LF seroprevalence was revealed among the sampled population, which could pose a serious public health threat. Intervention strategies should be implemented to check possible LF outbreaks in the sampled area, and further epidemiologic studies should be done to determine the means of spread and risk factors of Lassa fever.

Keywords: Lassa fever, Seroprevalence, River Banks, Immunoglobulin G, Mastomys natalensis

Significance | LF-specific IgG seroprevalence among inhabitants of river banks in Anambra State, Nigeria

*Correspondence: Chukwuma G.O., Department of Medical Laboratory Science, Nnamdi Azikiwe University Awka, Email. georgechuma@yahoo.com Phone. 08034101608

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Introduction

Lassa fever (LF) was discovered in 1969 in Nigeria but is endemic in many West African countries. About 300,000 to 500,000 cases of Lassa fever and 5000 deaths occur yearly across West Africa (Fitchet-Calvet et al., 2014; Ogbu et al., 2007). The illness was discovered in Lassa, Borno State, where it was first identified by three missionary nurses who died after caring for an obstetrical patient. Several epidemics have been recorded in many states in Nigeria with attendant high mortality rates. The Centre for Disease Control and Prevention (CDC) reported that Lassa

Author Affiliation:
[1] Department of Medical Laboratory Science, Nnamdi Azikiwe University Awka
[2] Molecular Research Laboratory, Nnamdi Azikiwe University Awka

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hemorrhagic fever caused by the Lassa virus is endemic in four countries in West Africa, including Guinea, Liberia, Sierra Leone, and parts of Nigeria (CDC, 2014). The host agent for the Lassa virus is the multimammate rat called Mastomys natalensis. A Lassa-infested rat does not become ill, but sheds the virus in its urine and droppings.

When humans come in direct contact with this urine, feces deposited on surfaces such as floors or beds, or in food or water, they become infected (WHO, 2005). Those who kill rodents for sport or eat rodents as deliccious are most likely to be infected by direct contact with its blood. Rat bites on humans is also another avenue for zoonotic transmission. Laboratory and person-to-person infections occur directly with blood and blood products, especially in the hospital environment. The incubation period for Lassa fever varies from 6 – 21 days. Lassa fever presents at its early stages with symptoms and signs indistinguishable from those of other viral, bacterial or parasitic infections common in the tropics such as malaria, typhoid and other viral haemorrhagic fevers such as Ebola (Tobin et al., 2013). Increased seasonal rainfall often leads to overflowing of banks of major rivers predisposing inhabitants of such areas to flooding and the consequent hazards that come with it. Flooding can lead to overcrowding of population and predisposition to unhygienic environmental conditions, poor sanitation, dilapidated or poor housing and proliferation of rodents that can now exacerbate the susceptibility of this population of people to Lassa fever infections among other diseases. This was affirmed by Clegg (2009) when he posited that "climate change is likely to lead to mass migration and movement of populations, with consequent stress associated with inadequate shelter and overcrowding. Such considerations are likely to be more significant concerning Lassa fever due to the much larger human population in the endemic area". It has also been established that the Lassa virus survives better in humid conditions during the rainy season, even though the viral aerosol stability seems to be higher when the humidity is lower, a condition that occurs more frequently in the dry season (Fichet-Calvet et al., 2008). This research was, therefore, an attempt to ascertain the extent of vulnerability of individuals who live in deplorable conditions along the banks of the River Niger to Lassa virus by determining their IgG seroprevalence. Few studies have been done on the IgG seroprevalence of Lassa fever virus among this population in South-Eastern Nigeria to the best our knowledge. Hopefully, this study's findings will expose the poor living conditions among individuals who reside on the river Niger bank.

Materials And Methods

Study Design

A cross-sectional study was conducted to determine the presence of Lassa fever antibodies in individuals who live in deplorable conditions along the banks of River Niger in Anambra State. A convenience sampling technique recruited subjects for the study.

Study Area

This study was carried out at Okpoko Community in Ogbaru LGA of Anambra state. Ogbaru is located between latitudes 6° 02' N and 6° 38' N and longitudes 06° 37' E and 06° 59' E. The average climatic conditions are wet (from March to October) and dry (from November to February) seasons. The average annual rainfall ranges between 1800 metres and 2000 metres. The temperature pattern has mean daily and annual temperature as 30°C and 27°C respectively, while the average relative humidity ranges between 60-70% and 80-90% in January and July respectively (Okoye et al., 2015). The Ogbaru LGA is a known flood-prone area due to its nearness to the River Niger and its low and flat topography with slope angles of 1°-3°. Thus, some communities are flooded for over 8 months yearly (Ajaero and Mozie, 2014). The Okpoko slum was selected for the study. The condition of buildings including the surrounding environment and other infrastructure in Okpoko slum is grossly uninhabitable and dangerous for human living (Okoye et al, 2017).

Study Population

A total of 90 subjects, both males and females, were recruited for this study, comprised of individuals who reside in Okpoko community, Ogbaru LGA, Anambra State.

Duration of Study

This study was carried out over three months, from August to October.

Ethical Consideration

Informed consent was sought and obtained from the subjects prior to data and sample collection. In addition, ethical approval for this study was obtained from the ethics committee of the College of Health Sciences, Nnamdi Azikiwe University by the Helsinki declaration by the World Medical Association on the ethical principles involving human subjects.

Data Collection

Data were collected using questionnaires given to the consenting respondents.

Specimen Collection

About 5 milliliters of venous blood was collected aseptically by a venepuncture from each consenting Subject and dispensed into sterile ethylene di-amine tetra-acetic acid (EDTA) anti-coagulant containers. Each specimen was labeled with the Subject's initials and laboratory identification number. Blood specimens were immediately transported to the laboratory and were separated by low-speed centrifugation at 500 x g for 5 minutes. The plasma was aseptically transferred into labeled sterile cryovials and stored at -20^{0}C until ready for analysis. Samples were analyzed at the

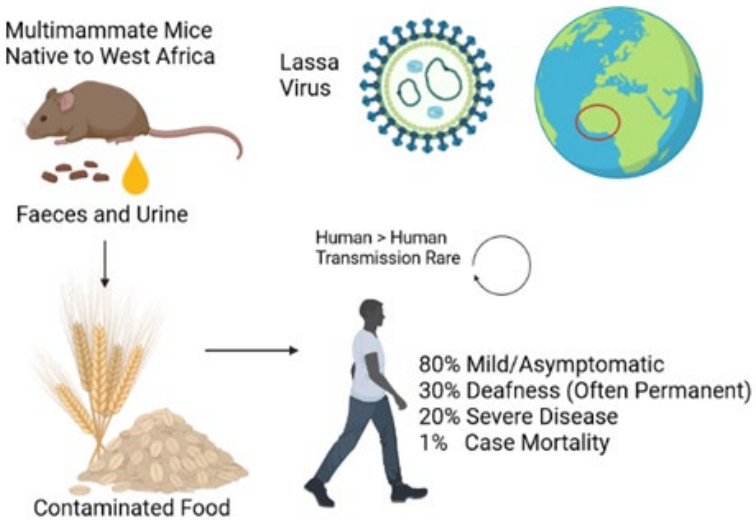

Figure 1. Transmission dynamics of Lassa fever virus in humans and rodents Culled from Lassa fever- *The Jenner Institute*

Table 1. Demographic characteristics of the selected study participants

Demographics	Frequency	Percentage (%)
Gender		
Male	18	20.0
Female	72	80.0
Total	90	100.0
Age (years)		
0-10	23	25.6
11-20	21	23.5
21-30	22	24.4
31-40	17	18.9
41-50	4	4.4
51-60	3	3.3
Total	90	100.0
Occupation		
Self-employed	54	60.0
Government-employed	11	12.2
Unemployed	25	27.8
Total	90	100.0
Level of education		
Primary	78	86.7
Secondary	12	13.3
Tertiary	0	0.0
Total	90	100.0
Marital status		
Married	50	55.6
Single	40	44.4
Total	90	100.0
Ethnic group		
Igbo	80	88.9
Hausa	3	3.3
Yoruba	2	2.2
Others	5	5.6
Total	90	100.0

-Molecular Research Laboratory, Nnamdi Azikiwe University Awka, Nnewi campus.

Method of Specimen Processing

The Enzyme-Linked Immuno-sorbent Assay (ELISA) technique was used to screen for Lassa fever-specific IgG antibodies in the samples. The ELISA kit was procured from Melsin Medical Company Limited China. The test was carried out according to manufacturer's instructions. The optical density (OD) was read at 450nm using a microtitre plate reader. The existence or not of Lassa Fever Virus IgG (LFV-IgG) in the samples is then determined by comparing the OD of the samples to the cut-off value determined by the manufacturer. A negative LFV result was interpreted as any sample with an optical density less than the calculated cut-off value and samples with an optical density greater than the calculated cut-off value were reported as positive for IgG to LF.

Data Analysis

The questionnaire results and data were analyzed with percentages and presented in tables. The statistical package for social science (SPSS) version 21, was used for data analysis. Simple prevalence and chi-square analysis were used where necessary and 95% level of significance at 0.05 confidence interval.

Result

Population characteristics of the study subjects

Females form the highest population, 72(80.0%). The 0-10 years is the highest occurring age range, 23(25.6), while the 51-60 age range was the least occurring, 3(3.3%). Most of the population were self-employed, 56(60.0%), and most stopped at the primary school level, 78(86.7%). Most of the subjects were married already, 50(55.6%), and most were from the Igbo extraction, 79(87.8%).

Prevalence of Lassa fever IgG among the population

The population with a negative outcome has the maximum value, 76(84.4%) while the population with a positive outcome has the lesser value, 14(15.6%). The negative population comprises of individuals with LF-specific IgG titre value less than the cut-off value (0.196), while subjects with a titre value greater than the cut-off value form the positive population.

Risk factors for Lassa fever infection.

Given the study area's environmental condition, the possible risk factors for Lassa fever infection were ascertained and its association with LF seropositivity was determined. In addition, each risk factor was correlated with the LF-specific IgG and their significance levels were determined.

Distribution of the association between LF seropositivity and age, gender and BMI

The sampled population with a normal BMI has the highest positive IgG titre value, 26(28.9%). The females have the highest occurrence in positive and negative IgG titre. The age range, 0-10 years has the highest occurrence in the negative population while the age range, 11-20 years and 21-30 years have equal positive LF-IgG occurrence. The p-values assuming the null hypothesis are insignificant since they are all higher than the alpha level (0.05). This means there is no statistical significance between LF seroprevalence against age, gender and BMI in this study.

Discussion

In this study, the Lassa fever-specific IgG was detected in 14 out of the 90 sampled individuals who are inhabitants of River Niger banks in Anambra State. This gave a prevalence rate of 15.6% . In the study of Kerneis et al. (2009), prevalence of positive LF immunoglobulin was 12.9% (10.8%-15.0%) and 10.0% (8.1%-11.9%) in rural and urban areas of Guinea, respectively. However, a study carried out in rural endemic Esan West local government area in Southern Nigeria showed seroprevalence at 58.2% where 96.1% of houses had seen rodents in the previous 6 months (Tobin et al., 2015). Although the prevalence in this study is not as high as that seen in Esan West LGA in Southern Nigeria, but the prevalence percent is sufficient to evoke a public health concern..

An assessment of the environmental and hygienic conditions of the community as seen shows that of the 90 subjects recruited for the study, 87(96.7%) persons see rats in their houses and 78(86.7%) individuals admitted that their environment is a very dirty one and unfit for human habitation. About 65(72.2%) persons affirmed no refuse-bin around their residence. However, 25(27.8%) subjects had open refuse bins in their area, and regrettably, 20(22.2%) admitted that these refuse bins are not being disposed of promptly. These, however, increase the level of indiscriminate refuse disposal, contributing to the unhygienic state of the area. Furthermore, 69(76.7%) subjects affirmed no portable toilet system. All of these encourage rodent's presence and persistence in human households, including the vector for LF. A correlation of these risk factors with Lassa fever seroprevalence rate, shows a considerable level of significance ($p \leq 0.05$), especially for persistence of rodents in households and poor refuse disposal. These findings are consistent with the assertions by some authors. Tambo et al. (2018) posits that Lassa fever is a known endemic infectious disease of poverty and has emerged as a severe outbreak of public health threat and burden in Nigeria in the recent past. There is increased risk in areas with poor-quality housing and in households with reduced levels of hygiene, poor sanitation and waste management (Bonner et al, 2007; Connolly, 2004). Most individuals are self-employed, where 54(60.0%), majorly petty traders and 25(27.8%) are unemployed. Most residents had only primary school education, 78(86.7%). Poverty and lack of adequate enlightenment invariably influence disease prevalence such as Lassa fever.

Table 2. Serological prevalence of Lassa fever virus among the study population.

LF IgG titre	Frequency	Percentage (%)
Negative	76	84.4
Positive	14	15.6
Total	90	100.0

Table 3. Association between seropositivity and risk factors of Lassa fever infection. ***Significant (p≤0.05)**

Possible risk factors	Lassa fever IgG		Total (%)	p-value
	Negative(%)	Positive(%)		
Rats in the house				
Yes	76(84.4)	11(12.2)	87(96.7)	0.000*
No	0(0.0)	3(3.3)	3(3.3)	
Total	76(84.4)	14(15.6)	90(100.0)	
Dirty environment				
Yes	10(11.1)	2(2.2)	12(13.3)	0.455
No	66(73.3)	12(13.3)	78(86.7)	
Total	76(84.4)	14(15.6)	90(100.0)	
Open refuse bins				
Yes	24(26.7)	1(1.1)	25(27.8)	0.031*
No	52(57.8)	13(14.4)	65(72.2)	
Total	76(84.4)	14(15.6)	90(100.0)	
Frequently disposed refuse				
Yes	4(4.4)	1(1.1)	5(5.6)	0.037*
No	7(7.8)	13(14.4)	20(22.2)	
Total	11(12.2)	14(15.6)	25(27.8)	
Portable toilet system				
Yes	17(18.9)	4(4.4)	21(23.3)	0.309
No	59(65.6)	10(11.1)	69(76.7)	
Total	76(84.4)	14(15.6)	90(100.0)	

Table 4. Association between LF seropositivity and age, gender and BMI.

Options	Negative (%)	Positive (%)	Total (%)	p-value
Age range(years)				
0-10	23(25.6)	0(0.0)	23(25.6)	0.221
11-20	16(17.8)	5(5.6)	21(23.3)	
21-30	17(18.9)	5(5.6)	22(24.4)	
31-40	14(15.6)	3(3.3)	17(18.9)	
41-50	3(3.3)	1(1.1)	4(4.4)	
51-60	3(3.3)	0(0.0)	3(3.3)	
Total	76(84.4)	14(15.6)	90(100.0)	
Gender				
Male	15(16.7)	3(3.3)	18(20.0)	0.885
Female	61(67.8)	11(12.2)	72(80.0)	
Total	76(84.4)	14(15.6)	90(100.0)	
BMI				
Underweight	20(22.2)	5(5.6)	25(27.8)	0.696
Normal weight	26(28.9)	5(5.6)	31(34.4)	
Overweight	15(16.7)	3(3.3)	18(20.0)	
Obesity	15(16.7)	1(1.1)	16(17.8)	
Total	76(84.4)	14(15.6)	90(100.0)	

Of the 90 subjects involved in the study, 18(20%) were males whereas the females were 72(80%). It can be seen from this study that seroprevalence was higher among females, 11(12.2%) than in males, 3(3.3%) . A study by Ilori et al. (2019) shows that girls and women accounted for a lower proportion of the laboratory-confirmed LF cases than boys and men (37.9% vs. 62.1%), but was not significantly different. A study by Kerneis et al. (2009) showed no significant difference in the seroprevalence of Lassa fever (LF) in females and males.

More so, for the age of the participants, it can also be seen that LF-specific IgG seroprevalence was higher in the following age brackets: 11-20 yrs, 5(5.6%) and 21-30 yrs, 5(5.6%) and was lowest in the following age ranges: 0-10 yrs, 0(0%) and 51-60 yrs, 0(0%). Kerneis et al. (2009), in their study showed a higher seroprevalence between 20 and 29 years of age and also highest attack rate (per 1000 persons) and LF antibody prevalence was seen in 20-29 years age bracket in a study conducted in Sierra Leone, 1970-1972 (Fraser et al, 1974). These agree with the findings in this study. In contrast, however, Fraser recorded a very low prevalence (least but one) in his study between 10-19 years of age which disagrees with the finding in this work as stated above. Studies by Ilori et al. (2019) and Fraser et al. (1974) reveal a significantly low seroprevalence in children between 0-10 years. This agrees with the findings in this study as no seropositivity was detected among this age group. But in the study by Kerneis et al. (2009), highest seroprevalence was seen in children below 10 years of age which contradicts the findings of Ilori et al. (2019), Fraser et al. (1974), and as well as results of this study. Ilori et al. (2019) reported a low prevalence in adults older than 50, which supports this study's findings.

On the contrary, study by Monath (1975) showed highest LF antibody prevalence among patients who are 40 years and above which is however not in tandem with our finding as very low seropositivity was detected in 41-50 yrs age range, 1(1.1%) and no seropositivity detected in 51-60 yrs age range. A graphical representation of the Lassa fever virus transmission dynamics is shown (Jenner Institute, 2022). LF virus is found in multimammate mice which are native to Nigeria and some West Africa states. It acts as reservoir of disease. The virus is transmitted to humans through contact with mice or their faeces and urine. This is usually through contaminated food. Human-to-human transmission is rare.

Conclusion

This study revealed a significant LF-specific IgG seroprevalence among inhabitants of river banks in Anambra State, Nigeria. This shows the percentage of riverine inhabitants who had previous exposure to LF virus. Unfortunately, this equally means that a greater percentage of residents on the River Niger bank are susceptible to infection with lassa fever virus. Currently, there are no vaccines to protect against infection. Therefore, a more aggressive campaign has to be carried out by the government and all stakeholders to improve the living conditions of riverine dwellers.

Author Contributions

U.K.C., C.G.O., C.O.M., designed research, U.K.C., C.G.O., C.O.M., N.I.M., performed research, U.K.C., C.O.M., O.M.B., M.P.O., N.I.M., C.G.O., analysed data, U.K.C., C.O.M., O.M.B., M.P.O., N.I.M., C.G.O., wrote paper.

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Competing financial interests

The authors have no conflict of interest.

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