Acta Scientific Clinical Case Reports

Volume 3 Issue 7 July 2022

Case Report

# Laboratory Diagnosis of Lassa Fever: Journey so Far in Nigeria

**Okeke Christiana Chiamaka[1]*, Tyem Nimnan Famak[1], , Edema Faith Modupe[1], Okeke Peace Nkiruka[2], Michael Eshioramhe Paul[3] and Olaniru Olumide[1]**

*[1]Jos University Teaching Hospital (JUTH), Nigeria*

*[2]Department of Microbiology, University of Jos, Nigeria*

*[3]Genomics and Postgraduate Research Laboratory, College of Health Sciences, University of Jos, Nigeria*

***Corresponding Author:** Okeke Christiana Chiamaka, Jos University Teaching Hospital (JUTH), Nigeria.

**Received:** May 30, 2022

**Published:** June 20, 2022



## Abstract

Lassa fever over the years has been endemic in West Africa, having its origin in the Northern parts of Nigeria. The continual occurrence of the disease in Nigeria most especially, has become an issue of serious concern. Modern methods to contain and control the spread of this deadly disease would require coordinated efforts by health practitioners as well as members of the community. Being that the major route of transmission is zoonotic, the elimination of the disease vector has always been the best bet in controlling the disease transmission. Further spread of the disease, can also be prevented through the adoption of proper environmental hygiene, appropriate decontamination of exposed surfaces and disposal of contaminated biological samples and waste. As a viral haemorrhagic fever, lassa fever is not easily distinguishable from other hemorrhagic fevers as the condition presents with similar signs and symptoms. Also, the occurrence of Lassa viral lineages has also contributed to making the differential diagnosis of the condition quite difficult. Therefore, the adoption of more advanced laboratory methods of diagnosis would go a long way to ensure more targeted and specific viral protein isolation and identification. This would also enable prompt responses, treatment and management of all suspected Lassa fever cases.

**Keywords**: Lassa Fever; Viral; Hemorrhagic; Laboratory; Diagnosis; Nigeria

## Introduction

Lassa fever, a viral hemorrhagic fever common to humans and primates was first discovered in 1969, when a missionary nurse named Laura Wine fell ill in Lassa, a village in Borno State, Nigeria. The cause of the disease was an enveloped, single-stranded, bisegmented, ambisense RNA virus known as the Lassa virus (LASV) [14]. The virus, *Lassa mammarenavirus*, was first isolated by Jordi Casals, from a fifty-two-year-old nurse named Lily Pinneo who attended to Laura [6]. The multimammate rat, *Mastomys natalensis*, was indicted as the main reservoir of the virus in West Africa, being able to shed the virus in its urine and feces without exhibiting visible symptoms [29]. Lassa viruses belong to the Arenaviridae family. Their genome is contained in two RNA segments, the large segment which encodes a small zinc protein (Z) that regulates transcription and replication, and the RNA polymerase (L) and the small segment which encodes the nucleoprotein (NP) and the surface glycoprotein precursor (GP), which is proteolytically cleaved into the envelope glycoproteins GP1 and GP2 that bind to the alpha-dystroglycan receptor and allow for host cell entry.

## History

Lassa fever garnered worldwide attention in 1969 when Missionary Nurses came down with a febrile illness of a mystic nature [6,7,14]. The nurses were believed to have acquired the infection while working in Lassa, a town located in Borno State,

North-Eastern Nigeria. In a quest for further treatment, the Nurses were conveyed to the Evangelical Church Winning All (ECWA) hospital in Jos. Two of the three nurses succumbed to death and the medical doctor who performed an autopsy on one of the nurses contracted the ailment and also died [6,7]. The only surviving nurse was taken to the United States of America where she was diagnosed with Lassa fever, she was subsequently treated and she survived. The Yale School of Medicine, Yale Arbovirus Unit was where Lassa fever was first isolated from the surviving nurse in 1970 [6,7].

## Epidemiology

Since the initial outbreak of Lassa fever, cases of the disease have continually been reported in Nigeria. The disease has also been recognized as being endemic in many parts of West Africa, including Nigeria, Benin, Ghana, Mali, and the Mano River region (Sierra Leone, Liberia, and Guinea). An estimated 300,000-500,000 cases and 5,000 related deaths occur annually in West Africa [8]. The prevalence of Lassa fever in Nigeria has been on the rise in recent times, with a higher proportion emanating from the southern part of the country. Lassa fever infections are usually associated with high mortality, especially in situations devoid of early access to diagnostic services and treatment. The epidemiology of Lassa fever in Nigeria bears semblance to that obtained in other countries within the West African sub-region [32]. Nigeria is endemic to Lassa fever with the peak occurring between April and December in the dry season period. 211 Lassa fever cases were confirmed by laboratory diagnosis in Nigeria between 3$^{rd}$ to 30$^{th}$ January 2022 with 40 deaths recorded within the same timeframe in 14 of the 36 states including the Federal Capital Territory. The majority of the cases accounting for about 82% were recorded in Ondo (63), Edo (57), and Bauchi (53). Other states with incidences are Benue (11), Ebonyi (5), Oyo (5), Taraba (5), Kogi (4), Enugu (2), Kaduna (2), Cross River (1), Delta (1), Katsina (1) and Plateau (1). An increase in the endemicity of Lassa fever was noted in 2022 from 2021, this is largely due to poor diagnostic services and reduced surveillance [38].

## Morbidity and mortality

Between the years 2015-2021, ages 21 to 40 years were the most infected by Lassa fever with a male to female ratio of 1:0.8. Of the 20,588 suspected cases of Lassa fever infections in Nigeria within the aforementioned period, only 3,311 cases were confirmed by laboratory diagnosis in 26 states with Plateau, Nassarawa, Edo, Taraba, Ondo, Bauchi, and Ebonyi recording yearly incidences. The case fatality rate ranged between 9.3% and 29.2% within this period. However, a decline has been observed in the epidemiology of Lassa fever infections between weeks 1 to 13 which are usually characterized by yearly seasonal peaks; a 75% reduction in Lassa fever incidence was observed between 2020 and 2021 [20].

## Disease vector

The multimammate rat which belongs to the family Muridae is scientifically known as *Mastomys natalensis,* it is the reservoir host of the Lassa fever Virus which is an arenavirus responsible for causing Lassa fever. The Multimammate rat is also known as the common African rat or African soft-furred mouse. The *Mastomys* rodent reproduces rapidly and is found in large numbers in the Savannah and forests of West, Central, and East Africa where they inhabit human homes and areas where food is readily available [11]. *Mastomys natalensis* is also a reservoir host to Mopeia virus in Southeastern Africa, Morogoro and Gairo virus in Tanzania and Luna Virus in Zambia; which are all arenaviruses. New hosts for the Lassa fever virus have been reported in Nigeria and Guinea they are *Mastomys erythroleucus* (Guinea multimammate mouse) and *Hylomyscus pamper* (African wood mouse) respectively [28].

## Transmission

Lassa virus (LASV) commonly spreads to humans by the natal multimammate mouse or African rat, (*Mastomys natalensis*). Other species of rats such as *Hylomyscus pamper* and *Mastomys erythroleucus* have been discovered as sources of the spread [28]. The means of Lassa fever transmission is mainly zoonotic through contact with either contaminated excreta or secretions. Once infected, the mouse excretes the virus throughout the rest of its lifetime through feces and urine. Handling and consumption of infected rodents is an infection pathway. The virus is transmitted by contact with the feces or urine of animals accessing grain stores in residences. The *Mastomys* rodents dwell in and around homes where they feed on food leftovers and not properly stored food in the process producing urine and excreta containing the Lassa fever virus. It is also possible to acquire the infection through broken skin or mucous membranes that are directly exposed to infectious material. Direct exposure to these materials containing the infectious agent either through touching soiled objects, consumption of contaminated food, or exposure to open cuts or sores can result in infection. Airborne transmission

may occur from aerosolized rodent excretions during cleaning activities, while transmission from person to person has also been established [29]. Human to Human transmission occurs either by exposure to the body fluids or secretions of an infected person. About 19% of all Lassa fever cases are transmitted by the human-to-human route, especially in hospital acquired infection settings [22]. Lassa fever can also be transmitted via sexual intercourse, due to the presence of Lassa fever nucleic acid in Semen. The virus is present in urine for between three and nine weeks after infection, and it can be transmitted in semen for up to three months after becoming infected [30]. The *Mastomys* rodents sometimes serve as a food source, a common practice in some parts of Nigeria, Lassa fever infection can therefore be transmitted when these rodents are consumed [11]. Lassa fever virus transmission is primarily by environmental exposure. This is in contrast to that of the Ebola virus which is by prolonged human to human transmission [3].

## Pathogenesis

The defining feature of Lassa fever pathogenesis is the inability of the immune system to mount a response due to the inactivation of the immune cells responsible for innate immunity (dendritic cells and macrophages). A high viral load is a common finding in severe Lassa fever episodes. Interferon responses are blocked by the small matrix proteins and nucleoproteins of the Lassa fever particle this is achieved by halting viral RNA sensing thus preventing antigen presentation and T-cell responses [12]. A strong and early T cell immune response is an indicator of Lassa fever survival than a B cell immune response. The incubation period of Lassa fever is between 7 to 21 days [2]. Upon entry into the human body, the Lassa virus first infects the mucosa, then the intestine, the lungs, the urinary system, and finally the vascular system targeting mainly antigen-presenting cells (Donaldson, 2009). The host innate defense system recognizes the Pathogen-associated molecular patterns (PAMP) and activates an immune response. One of the mechanisms is the detection of double-stranded RNA (dsRNA) synthesized by negative-sense viruses by dsRNA receptors, such as RIG-I (retinoic acid-inducible gene I) and MDA-5 (melanoma differentiation associated gene 5). These receptors initiate signaling pathways that translocate IRF-3 (interferon regulatory factor 3) and other transcription factors to the nucleus. Translocated transcription factors activate the expression of interferons α and β, initiating adaptive immunity. The Lassa Virus Nucleoprotein (NP) essential in viral replication and transcription is also said to have an exonuclease activity to only dsRNAs which allows it to suppress host innate IFN response by inhibiting translocation of IRF-3 [18]. The NP dsRNA exonuclease activity counteracts IFN responses by digesting the PAMPs thus allowing the virus to evade host immune responses.

## Signs and symptoms

After an incubation period of 7 to 21 days, symptoms begin in those who are infected while few or no symptoms may also occur. In cases in which death occurs, this typically occurs within 14 days of the onset of infection [8]. A combination of pharyngitis, pain behind the sternum, presence of excess protein in the urine, and fever can be used to distinguish Lassa fever from other hemorrhagic fevers with higher specificity (McCormick, 1987). Signs and symptoms of Lassa fever infection persist for up to 3 weeks after the infection is acquired [1]. Lassa fever infection in humans can result in mild infection in about 80% of all cases thus prompting most Lassa fever cases to remain undiagnosed and undetected in certain populations [1,14]. Initial disease manifestation is usually gradual, accompanied by non-specific symptoms such as general weakness, malaise, fever, and headache. The disease then progresses after a few days, where symptoms worsen with a sore throat, chest pain, muscle pain, vomiting, diarrhea, arthralgia, cough, and nausea; these symptoms may be followed by pain in the back and abdomen. The disease may then progress to more serious symptoms in one out of five infected individuals. Severe cases are characterized by severe bruising, facial swelling, petechiae, hepatitis, renal failure, respiratory distress, fits, tremors, gait disturbance, disorientation, loss of consciousness, bleeding from the mouth, and nose, gastrointestinal tract, or vagina. Bleeding is a common finding in about 30% of all Lassa fever cases [1,14]. Varying degrees of deafness occur in about one-third of all cases and most times result in permanent hearing loss. Notwithstanding the severity of the disease, deafness may occur in mild and severe Lassa fever cases [11].

## Specimen collection and handling

Due to the high risk posed by Lassa fever spread via body fluids, medical laboratory personnel should be wary of the risk of handling potentially infectious Lassa fever specimens since all specimens are regarded as being potentially infectious. Laboratory personnel may be exposed to safety hazards due to poor storage and handling of

Lassa fever specimens which may result in decreased sensitivity of the diagnostic assays employed [31]. When caring for patients with suspected or confirmed Lassa fever cases health workers should employ extra infection control measures to prevent contact with the patient's body fluids and contaminated surfaces or materials such as clothing and bedding. Health workers should put on face protection equipment, a clean long sleeve gown, and sterile gloves when coming in close contact (within 1 meter) with suspected Lassa fever patients to perform procedures such as sample collection [31]. When handling specimens which may contain the Lassa fever virus, Biosafety Level-4 precautions are recommended. However, in the absence of Biosafety Level-4 laboratories, samples for Lassa fever diagnosis can be handled in Class II or III biosafety cabinets. Alternatively, samples may be inactivated to allow for safe handling of specimens under Biosafety Level-2 precautions [31].

## Laboratory diagnosis

Nigeria has seven national Lassa fever laboratories managed by the National Center for Disease Control that can conduct molecular testing for Lassa fever, these laboratories operate at full capacity to improve turnaround time [31]. Lassa fever symptoms are non-specific and vary greatly, making clinical diagnosis very difficult, especially early in the course of the disease. Most febrile illnesses such as malaria, dengue, influenza, and yellow fever which are endemic to the West African sub-region present with signs and symptoms similar to Lassa fever, hence differentiating between their agents remains a major obstacle [31]. Most times, self-treatment for malaria or bacterial infection is performed and Lassa fever comes into the foray only after the patient fails to recuperate following antimalarial or antibiotic therapy. Therefore, laboratory diagnosis is essential for the confirmation of Lassa fever disease. There are at least five known lineages (Strains I - V) of LASV with a Sixth lineage being proposed. Because of the vast lineage diversity and frequency of mutations, there has been much need for advancements in laboratory methods of diagnosis to encompass variations observed at the viral nucleotide and amino acid levels. According to Nigeria's National guidelines for Lassa fever case management, investigations to be performed for the diagnosis of Lassa fever are the Reverse Transcriptase Polymerase Chain Reaction (RT-PCR) and serological assays encompassing Rapid Diagnostic Testing (RDT). In case scenarios where the illness is mild, supportive investigations comprising of full blood count, liver function tests, urinalysis, malaria parasite test and pregnancy test for women of all childbearing age should be conducted. While in severe illness all the investigations for mild Lassa fever illness should be conducted along with Electrolyte, Urea, and Creatinine estimation; random blood glucose and quantitative Polymerase chain reaction for monitoring purposes, blood culture, prothrombin time, blood gases, serum calcium, and based on clinical suspicion others tests may be performed [8]. For the laboratory diagnosis of Lassa fever in Nigeria, the following methods are employed.

## Virus isolation by cell culture

Viral culture is the gold standard for LASV isolation, however, it must be performed in laboratories with good containment practices. The Lassa fever virus can be cultured between 7 to 10 days. Formalin fixed tissues can be utilized to perform immunohistochemistry to make a post mortem diagnosis [9].

## Indirect immunofluorescence assay test (IFA or IIFT)

The traditional method for LASV isolation and serodiagnosis was the Immunofluorescence assay test using infected Vero cells. However, this has been largely displaced by ELISA due to time and biosafety constraints [31]. In the past, Immunofluorescence assay (IFA) was used to detect LASV antibodies. However, the requirements for BSL-4 biocontainment, highly trained technicians, and inferior level of sensitivity have limited its use.

## Western blot (WB)

Western blot is often used to separate and identify proteins. A mixture of proteins is separated based on molecular weight and type, through gel electrophoresis. These results are then transferred to a membrane producing a band for each protein. The membrane is then incubated with labels antibodies specific to the protein of interest. The unbound antibody is washed off leaving only the bound antibody to the protein of interest. The bound antibodies are then detected by developing the film. As the antibodies only bind to the protein of interest, only one band should be visible. The thickness of the band corresponds to the amount of protein present, thus, doing a standard can indicate the amount of protein present [5].

## Rapid diagnostic test (RDT)

LASV RDTs, uses the same antibody/antigen capture agents as an ELISA but packaged in a stripped-down lateral flow format,

detect NP from fingerstick whole blood specimens [5]. In low- and middle-income countries such as Nigeria which are often characterized by a lack of technological wherewithal and epileptic power supply, assays such as RT-PCR and ELISA are difficult to perform. Presently, only one Lateral Flow Immunoassay method used for point of care testing for the detection of Lassa fever is available, this is the ReLASV Rapid Diagnostic Test (RDT) which is a dip stick methothatch that makes use of paired Lassa fever virus nucleoprotein mouse monoclonal antibodies produced from recombinant nucleoprotein immunized mice. Detection of the Josiah strain of Lassa virus prevalent in Sierra Leone was the primary reason why this method was developed [5]. For the ReLASV Pan-Lassa Antigen Rapid Test, the LASV NP-specific antibody is bound to a nitrocellulose strip to capture the LASV NP antigen. On one of the rapid tests, reagent pads lie gold nanoparticles conjugated to Lassa fever virus-specific antibody. During the assay, the Lassa fever nucleoprotein antigen in the sample produces immune complexes with the anti-LASV NP antibody test strip, thus producing a pink to red color which is directly proportional to the concentration of Lassa fever virus NP antigen in the sample. The anti-rabbit IgG control strip captures the excess gold-conjugate which is indicative of a valid result. The result is interpreted visually between 15 to 25 minutes of color development [39]. While LFIs can be an important part of an orthogonal system, they suffer from poor sensitivity and specificity compared to other methods. They should never be used as a standalone diagnostic, but when used judiciously, can improve the overall diagnostic process and outcome.

## Enzyme-linked immunosorbent assay (ELISA)

This is a relatively simple to perform an assay that can be conducted in low- and middle-income countries where Lassa fever is endemic. The method is used to detect viral proteins or LASV-specific IgM or IgG antibodies [26]. Detection of antigen generally employs a capture or sandwich ELISA. A "capture" antibody conjugated to solid support binds LASV in a sample. After washing away non-binding components, the captured antigen is "sandwiched" by a primary antibody. The addition of a secondary (anti-species-specific) antibody conjugated to a reporter molecule produces a detectable signal. IgM, the first and thus most clinically relevant antibody produced after infection, can be detected using sandwich ELISA. Detection of the Lassa fever antigen or IgM-specific antibodies is important in making a Lassa fever diagnosis. IgG antibodies are produced later during the infection hence detection of Lassa fever-specific IgG antibodies has little or no diagnostic significance. This method of Lassa fever diagnosis is commonly used for disease surveillance. The Sandwich ELISA method is performed in a quest to detect an antigen, for this assay, an antibody is known as the primary antibody is coated unto the wall of a solid support membrane such as a microtitre plate or a cellulose membrane to bind the Lassa fever antigen in the sample under investigation. The reaction mixture is then washed to remove unbounded components. A secondary antibody that is anti-species-specific and attached to a chromogen is added to elicit a detectable signal. Using sandwich ELISA, IgM antibody which is usually the first antibody produced in response to an infection can be detected. This is achieved by adding a serum sample containing Lassa fever IgM antibodies to the reaction mixture already containing the Lassa fever antigen bound to the primary antibody. An anti-species-specific antibody conjugated to a chromogen is then added to emit a detection signal. ELISA for detection of either the antigen or IgM antibody requires whole inactivated virus or virus infected cell lysate to serve as the positive control [17].

## Magnetic bead-based immunoassay

This uses magnetic beads with unique fluorescent labels resulting from different ratios of two fluorophores. Assays performed are similar to ELISA, but with a greater advantage as the magnetic beads serve as the solid support structure for antibodies. The development of a magnetic bead-based immunoassay using antigens and IgM for Lassa and Ebola has helped to enhance differential diagnosis by demonstrating greater detection sensitivity for LASV GP antigen and IgM than conventional ELISA [33].

## Reverse transcriptase -PCR (RT-PCR)

In the early 2000s Lassa fever cases were diagnosed and confirmed from far and wide within Nigeria at the Virology Unit Laboratory, College of Medicine, University of Lagos (CMUL) using the Reverse Transcriptase Polymerase Chain Reaction (RT-PCR) [37]. Reverse Transcriptase-polymerase chain reaction (RT-PCR) is recommended for use during the early stages of Lassa fever disease. It is designed to detect highly conserved regions of the pathogen genome (LASV RNA) and is commonly used as the most sensitive method to detect active infection. In RT-PCR, the RNA template is first converted into a complementary DNA (cDNA) using reverse transcriptase (RT). The cDNA is then used as a template

for exponential amplification using PCR. The quantification of mRNA using RT-PCR can be achieved as either a one-step or a two-step reaction. The difference between the two approaches lies in the number of tubes used when performing the procedure. The two-step reaction requires that the reverse transcriptase reaction and PCR amplification be performed in separate tubes. The disadvantage of the two-step approach is susceptibility to contamination due to more frequent sample handling [37]. The one-step approach minimizes experimental variation by containing all of the enzymatic reactions in a single environment. It eliminates the steps of pipetting cDNA products, which is labor-intensive and prone to contamination [21]. However, the starting RNA templates are prone to degradation in the one-step approach, and the use of this approach is not recommended when repeated assays from the same sample are required. Additionally, the one-step approach is reported to be less accurate compared to the two-step approach.

### Real time reverse transcriptase PCR (RT-qPCR)

Quantitative PCR (qPCR) is used to detect, characterize and quantify nucleic acids for numerous applications. Commonly, in RT-qPCR, RNA transcripts are quantified by reverse transcribing them into cDNA first, as described above and then qPCR is subsequently carried out. However, in qPCR, fluorescent labeling enables the collection of data as PCR progresses. The combination of RT-PCR with tagged primers can enable the detection of distinct LASV species and this has helped to overcome the limitations of fluorescence detection. Others have paired conventional PCR assays with multiplex detection formats such as magnetic bead-based platforms and microarrays, which serve as the solid support structures for oligonucleotides thereby improving sensitivity. Real-Time Reverse Transcriptase Polymerase Chain Reaction (RT-PCR) has the unique feature of being highly sensitive and specific, hence it has become the standard and is a common diagnostic and confirmatory tool for Lassa fever. Samples can be processed in large quantities in good time and at much cheaper rates using 96 well-plate-based thermocyclers, polymerase chain reaction has the advantage over the viral culture of being able to detect Lassa fever for a longer period and during early episodes of the illness [36]. PCR can also be performed on specimens that have been chemically inactivated. The 95% probability limit using RT-PCR varies depending on the virus strain and primers employed in the process, the estimate varies from between 1,237 to 4,290 RNA copies/ml. Due to the highly diverse nature of the Lassa fever virus, nucleic acid-based assays are faced with the issue of genetic diversity, as a primer with a single nucleotide variant can exert a significant negative impact on the assay's sensitivity depending on where the nucleotide variant is located [34].

### Next generation sequencing (NGS)

This is also known as massively parallel sequencing and it refers to a high technology reliant assay that obtains important information from a genome in an efficient and timely manner. The emergence of NGS has impacted the field of biological science, enabling researchers to probe further to obtain information about the genome. NGS uses metagenomics to discover new viruses, monitor outbreaks before they get out of hand, understand diversity and provide guidance for management to make decisions [35]. NGS and other molecular techniques point to the fact that Lassa fever has been in existence in Nigeria for 1000 years and between 150 to 400 years in parts of West Africa and Sierra Leone, but it was only in 1979 that Lassa fever was discovered in Nigeria. Real-time NGS data was utilized for the management and control of the Lassa fever outbreak that occurred in 2018 thus dousing fears that arose due to an increase in the prevalence of Lassa fever. The NGS data pointed to the fact that transmission was majorly from the vector *Mastomys natalensis* with little human-to-human transmission apart from nosocomial cases [23].

### Clinical management

Convalescent plasma has been found useful in the management of Lassa fever disease as plasma from a patient who previously had the infection is introduced into a patient with an active Lassa fever infection. Convalescent plasma proved useful in the management of the Argentine hemorrhagic fever caused by the Junin virus also an arenavirus. Patients who receive convalescent plasma recuperate faster than patients who do not [15].

### Treatment

Ribavirin is a guanosine analog that exerts a virus-static effect on a large number of viruses [16]. The mode of action of Ribavirin on Lassa fever virus is not certain but it has been reported to act on viruses with different mechanisms, these mechanisms include inhibition of viral RNA-dependent RNA polymerases, the inhibition of viral capping enzymes, and the inhibition of host inosine monophosphate dehydrogenase (IMDPH) [6]. During the 2018 outbreak of Lassa fever in Nigeria, an estimated 1,893

cases were identified with only 423 confirmed by laboratory diagnosis. 94.1% of the patients with confirmed Lassa fever cases were treated with Ribavirin. The case fatality rate of those who were treated with Ribavirin was 20.7% compared with 71.4% of those who did not receive treatment with the drug [19]. During the study the patients were put into three categories: the lowest fatality rate was observed in those who received treatment with Ribavirin within seven days from the onset; patients who received treatment with Ribavirin more than seven days from the onset and those who did not receive treatment with Ribavirin at all. It was observed that patients who received treatment with Ribavirin after seven days from the onset had a lower-case fatality rate than those who did not receive treatment with Ribavirin, at all [19]. For average-weight adults, the recommended dosing for intravenous administration of Ribavirin is a 2.4g loading dose, to be followed by administration of 1g every 6 hours for 10 days. Oral Ribavirin is used for post-exposure prophylaxis at a dose of 500-600 mg every 6 hours for a period of between 7 to 10 days. Dose adjustments are made for patients with a creatinine clearance rate of above 50 ml/min [24]. The main side effect of Ribavirin is hemolysis which is dose dependent and occurs in approximately 20 percent of patients. Other side effects of oral treatment with ribavirin include nausea, vomiting, diarrhea, myalgia, dry mouth, fatigue, headache, rash, jaundice, tachycardia, thrombocytosis, increased lipase levels, mood changes, and insomnia [4].

## Surveillance and disease control

Improved surveillance for Lassa fever is ongoing in all states of Nigeria. This encompasses increased case findings. Data garnered across the country is used to inform situational updates by Nigeria's Center for Disease Control and set a framework for further response plans [38]. Surveillance is carried out in Nigeria via the Integrated Disease Surveillance and Response (IDSR) platform. Data on Lassa fever from health facilities is forwarded to the Local Government Area (LGA) Disease Surveillance and Notification Officers (DSNOs) via the ward focal persons and from there to the State epidemiologist and ultimately to Nigeria's Center for Disease Control and Federal Ministry of Health [27]. By preventing contact with the vector *Mastomys natalensis*, transmission from the host to humans will be halted especially in regions where Lassa fever outbreaks occur. Hygienic practices such as storing food in rodent-proof containers and keeping homes clean are means of preventing the infection. It is advised to avoid the consumption of the vector rodent. Precautions such as wearing protective clothing, masks, gloves, gowns, and Google go a long way in preventing person to person or even nosocomial transmission of Lassa fever. Effective sterilization of hospital equipment and isolation of patients infected with the Lassa virus disease until the disease has run its course are other means of controlling the disease spread. Engagement in public health enlightenment programs in areas prone to Lassa fever outbreaks on measures of decreasing rodent populations in their homes will prove valuable in a bid to prevent and control Lassa fever outbreaks. Also, more rapid diagnostic test methods and other options for treatment could assist in the control of Lassa fever in Nigeria [10].

## Vaccines

Currently, there is no approved vaccine for Lassa fever. However, a vaccine has been developed although still under investigation by scientists at Thomas Jefferson University, Philadelphia; University of Minho, Braga Portugal; the University of California, San Diego and the National Institute of Allergy and Infectious Diseases (NIAID) and part of the National Institutes of Health. The vaccine called LASSARAB is aimed at conferring immune protection for both rabies and Lassa fever and it is a recombinant vaccine candidate which utilizes a weakened rabies vector or carrier. Genetic material from the Lassa virus was put into the vector for the rabies virus to enable the vaccine express surface proteins from both Rabies virus and Lassa virus. An immune response is then triggered by the surface proteins against both Lassa fever and rabies viruses. The recombinant vaccine was then attenuated by killing the live Rabies virus which was utilized in producing the carrier [25].

## Community perspective

There have been community mobilization and sensitization efforts toward Lassa fever prevention and control in Africa. Also in a bid to prevent Lassa fever transmission, rodent control practices, environmental sanitation, proper food processing and storage, and stakeholders' advocacy have been initiated. A community to health facility referral mechanism has also been initiated through the engagement of health workers [19].

## Conclusion

In the fight against Lassa fever, laboratory diagnosis is one of the most effective tools being employed. Therefore, the gradual

advancements noted in this area give a clear indication that in the no distant future, the challenges faced in the management and treatment of this disease condition will be a thing of the past in Nigeria and West Africa.

## Bibliography

1. Akpede G., *et al*. "Caseload and Case Fatality of Lassa Fever in Nigeria, 2001-2018: A specialist Center's Experience and its implications". *Frontiers in Public Health* 7 (2019): 251.

2. Asogun, D., *et al*. "Lassa Fever-Epidemiology, Clinical Features, Diagnosis, Management, and Prevention". *Infectious Disease Clinics of North America* 6 (2019).

3. Baize S., *et al*. "The emergence of Zaire Ebola Virus Disease in Guinea". *New England Journal of Medicine* 371 (2014): 1418-1425.

4. Bausch D., *et al*. "Review of the literature and proposed guidelines for the use of oral ribavirin as post-exposure prophylaxis for Lassa fever". *Clinical Infectious Diseases* 51 (2010): 1435-1441.

5. Boisen ML., *et al*. "Field validation of recombinant antigen immunoassays for diagnosis of Lassa fever". *Science Representation* 8 (2018): 5939.

6. Buckley S., *et al*. "A new virus disease of man from West Africa III Isolation and Characterization of the virus". *American Journal of Tropical Medicine and Hygiene* 19 (1970): 670-676.

7. Carey D., *et al*. "Epidemiological aspects of the 1970 epidemic, Jos, Nigeria". *Transactions of the Royal Society of Tropical Medicine and Hygiene* 66.3 (1972): 402-408.

8. CDC "Signs and Symptoms, Lassa Fever" (2019).

9. Center for Disease Control and Prevention. Lassa fever-Diagnosis (2014).

10. Center for Disease Control and Prevention. Lassa fever-Prevention. US Department of Health and Human Services (2014).

11. Center for Disease Control and Prevention. Lassa Fever. Transmission (2014).

12. Cohen-Davshi H., *et al*. "Molecular Mechanism for LAMP1 recognition by Lassa virus". *Journal of Virology* 89 (2015): 7584-7592.

13. Drosten C., *et al*. "Rapid detection and quantification of RNA of Ebola and Marburg viruses, Lassa virus, Crimean-Congo hemorrhagic fever virus, Rift valley virus, dengue virus, and yellow fever virus by real-time reverse transcription-PCR". *Journal of Clinical Microbiology* 40 (2002): 2323-2330.

14. Frame J., *et al*. "Lassa fever is a new virus disease of man from West Africa. I. Clinical description and pathological findings". *American Journal of Tropical and Medical Hygiene* 19.4 (1970): 670-676.

15. Frame J., *et al*. "The use of Lassa fever convalescent plasma in Nigeria". *Transactions of the Royal Society of Tropical Medicine and Hygiene* 78 (1984): 319-324.

16. Gowen B and Bray M. "Progress in the experimental therapy of severe arenaviral infections". *Future Microbiology* 6 (2011): 1429-1441.

17. Happi A., *et al*. "Lassa Fever Diagnostics: Past, Present, and Future". *Current Opinion in Virology* (2019).

18. Hastie KM., *et al*. "Structural basis for the dsRNA specificity of the Lassa virus NP exonuclease". *PLOS ONE* 7.8 (2012): e44211.

19. Ilesanmi O., *et al*. "Community engagement in the prevention and control of Lassa fever in Africa: A Systematic Review". *Infection Epidemiology and Microbiology* 7.2 (2021): 187-196.

20. John-Ugwuanya A., *et al*. "Epidemiological trends of Lassa fever in Nigeria from 2015-2021: A review". *Therapeutic Advances in Infectious Disease* 8 (2021): 1-7.

21. Li L., *et al*. "Development of a direct reverse-transcription quantitative PCR (dirty-qPCR) assay for clinical Zika diagnosis". *International Journal of Infectious Diseases* 85 (2019): 167-174.

22. Lo Lacono G., *et al*. "Using modeling to disentangle the relative contributions of zoonotic and anthroponotic transmission: the case of Lassa fever". *PLOS Neglected Tropical Diseases* 9 (2015).

23. Manning J., *et al*. "Lassa virus isolates from Mali and the Ivory coast represent an emerging fifth lineage". *Frontiers in Microbiology* 6 (2015): 1037.

24. McCormick J., *et al*. "Lassa fever: Effective therapy with ribavirin". *New England Journal of Medicine* 314 (1986): 20-26.

25. National Institutes of Health. "Scientists develop a novel vaccine for Lassa fever and rabies". NIH Research Information (2018).

26. Niedrig M., *et al*. "First international quality assurance study on the rapid detection of viral agents of bioterrorism". *Journal of Clinical Microbiology* 42 (2004): 1753-1755.

27. Okoro O., *et al*. "Descriptive epidemiology of Lassa fever in Nigeria, 2012-2017". *Pan African Medical Journal* 37.15 (2020): 1-7.

28. Olayemi A., *et al*. "Arenavirus Diversity and Phylogeography of Mastomys natalensis Rodents, Nigeria". *Emerging Infectious Diseases* 22.4 (2016): 694-697.

29. Patassi AA., *et al*. "Emergence of Lassa Fever Disease in Northern Togo: Report of Two Cases in Oti District in 2016". *Case Repeat of Infectious Disease* (2017): 1-5.

30. Public Health England. "Lassa fever: origins, reservoirs, transmission, and guidelines" (2014).

31. Raabe V and Koehler J. "Laboratory diagnosis of Lassa fever". *Journal of Clinical Microbiology* 55 (2017): 1629-1637.

32. Saka J., *et al*. "Lassa Fever Epidemic in Nigeria- Outbreak Investigation, Risk Factors and Empirical Analysis from 2012 to 2016". *Journal of Community and Public Health Nursing* 3 (2017): 170.

33. Satterly NG., *et al*. "Comparison of MagPix Assays and Enzyme-Linked Immunosorbent Assay for Detection of Hemorrhagic Fever Viruses". *Journal of Clinical Microbiology* 55 (2017): 68-78.

34. Stadhouders R., *et al*. "The effect of primer-template mismatches on the detection and quantification of nucleic acids using the 5' nuclease assay". *Journal of Molecular Diagnosis* 12 (2010): 109-117.

35. Strenglein M., *et al*. "Identification, characterization, and in vitro culture of highly divergent arenaviruses from boa constrictors and annulated tree boas: candidate etiological agents for snake inclusion body disease". *Molecular Biology* 3 (2012): 00180-00112.

36. Trappier S., *et al*. "Evaluation of the polymerase chain reaction for diagnosis of Lassa virus infection". *American Journal of Tropical Medicine and Hygiene* 49 (1993): 214-221.

37. Wong ML and Medrano JF. "Real-time PCR for mRNA quantitation".*BioTechniques* 39.1 (2005): 75-85.

38. World Health Organization. "Lassa Fever-Nigeria". Disease Outbreak News (2022).

39. Zalgen. "ReLASV Pan-Lassa Antigen Rapid Test RUO (10376)". Zalgen Labs (2020).