Research Article

# Prevalence and Genomic Characterization of Zika Virus among Pregnant Women in Plateau State, Nigeria

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**Abstract** | This study aimed to investigate the prevalence and molecular characterization of Zika virus (ZIKV) infection among pregnant women attending prenatal clinics in Nigeria. Since ZIKV, a flavivirus primarily transmitted by Aedes mosquitoes, is associated with neurological complications and congenital defects, it poses a significant public health threat. A total of 324 consenting pregnant women from the three senatorial zones of Plateau State participated in a cross-sectional study. The presence of ZIKV was determined using one-step real-time RT-PCR after extracting total RNA from sera and urine samples. Positive samples were further characterized with conventional RT-PCR and Sanger's sequencing. Overall, 6.8% (22/324) of urine samples and 4.9% (16/324) of serum samples tested positive for ZIKV RNA, indicating that urine is more sensitive for ZIKV RNA detection. Molecular characterization revealed nine isolates belonging to the Asian genotype. Notably, one isolate was identified as part of the American Asian Bridge lineage (Mixed USA-Singapore), while seven isolates belonged to the American lineage (including two Mexican and four American/USA sub-lineages). No evidence of an African genotype was found. These findings demonstrate that imported ZIKV strains are circulating in Plateau State, highlighting the importance of robust monitoring systems especially for pregnant women to track the introduction and spread of different ZIKV genotypes and implement targeted preventive measures to reduce the risk of adverse pregnancy outcomes.

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## Introduction

Zika virus (ZIKV) is a positive-sense, single-stranded RNA virus of the genus Flavivirus, family *Flaviviridae*. ZIKV is an arthropod-borne virus, primarily transmitted to humans by the bite of an infected Aedes mosquito, specifically of the subgenus Stegomyia (Cirne-Santos, 2024; Tajik *et al.*,

2024). Sexual contact, intrauterine transfer, perinatal transmission, laboratory exposure, and perhaps blood transfusion are additional transmission pathways (CDC, 2024; Dawurung *et al*., 2025). In the general population, ZIKV infection is usually asymptomatic; however, it can present with modest symptoms such as fever, arthralgia, non-purulent conjunctivitis, and maculopapular rashes (Pielnaa *et al*., 2020; Dappa and Ogbonnaya, 2023).

Infection in pregnancy can present with serious negative outcomes; these include miscarriage, Guillain-Barré syndrome, congenital microcephaly, various brain disorders, and stillbirth (Beber *et al*., 2023; Dawurung *et al*., 2025; Pérez *et al*., 2025). The congenital Zika syndrome, which is the aggregate term for severe fetal problems, emphasizes its vital public health significance, particularly for expectant mothers (Pérez *et al*., 2025); thus, it is considered a serious public health issue of global concern.

In Nigeria, ZIKV infection was first reported in 1975 (Diallo *et al*., 2023; Moore *et al*., 1975), and since then, researchers have continued to share their findings. A study conducted between 1971 and 1975 in Oyo State reported a sero-prevalence of 31% (Fagbami, 1979). Similarly, Carteaux and colleagues conducted a study in North-Central Nigeria and reported a prevalence of 6% and 4%, for IgM and IgG, respectively (Carteaux *et al*., 2016). More recently, a cross-sectional study from Plateau State found a seroprevalence of 14.4% (Anejo-Okopi *et al*., 2020).

It is therefore obvious that there still exists dearth of comprehensive information on ZIKV infection in Nigeria. Understanding the genetic composition of the circulating strains, spotting transmission patterns, and precisely estimating the current situation of ZIKV infection in the country are crucial for future epidemiological and clinical decisions. This current study investigated ZIKV infection among pregnant women at various stages of pregnancy across the three senatorial zones in Plateau State and explored the genetic diversity of the circulating strains of the virus in the study area with a view to bridging this important information gap.

## Materials and Methods

### *Study area/population*

We conducted the study in nine (n=9) public health facilities among pregnant women in Plateau State, the state is located approximately in the center of Nigeria, Plateau State is geographically distinctive, characterized by its elevated hills surrounding the Jos Plateau (Iro *et al*., 2019; PSG, 2023). Plateau state shares boundaries with 4 states and has a unique temperate weather which attracts tourists, has a mean annual rainfall of 1460 mm. The state which is the gate way to the Northeastern states from the south. The state is well known of its agricultural activities because of it's fertile soil and favourable climate for agricultural activities. Plateau state is multiethnic, cosmopolitan, and headquarters to several federal agencies in Nigeria. The state also has an international airport and dryland port which encourages travels both locally and international. The selected hospitals were: Northern Zone: General Hospital Barkin Ladi (Lat. 9.534289, Long. 8.907348), Comprehensive Health Centre Dadin Kowa (Lat: 9.851294, Long: 8.866973), and Primary Health Centre Rayfield (Lat: 9.830752, Long: 8.889400); Central Zone: General Hospital Mangu (Lat: 9.475262, Long: 9.161370), Cottage Hospital Bokkos (Lat: 9.295800, Long: 8.979266), and Township Primary Health Centre, Pankshin (Lat: 9.324302, Long: 9.438680); Southern Zone: General Hospital Shendam (Lat: 8.872728, Long: 9.532859), Cottage Hospital Kwalla (Lat: 8.924814, Long: 9.282970), and Primary Health Centre Shendam A (Lat: 8.878122, Long: 9.540806).

### *Study design and sampling technique*

This study was cross-sectional in design. Participants were selected using a simple random method following a series of group health education and enlightenment sessions.

### *Sample size determination*

The sample size for the study was determined using the formula put forward by Lwanga and Lemeshow (1991). A total of 324 participants were recruited, making up of 108 per senatorial zone.

### *Inclusion and exclusion criteria*

All pregnant women who consented to participate in the study were recruited, and women who were pregnant and attended the clinics during the study period but denied consent were excluded.

### *Informed consent*

Before the commencement of data collection, informed consent was obtained from each of the participants.

*Data collection*

The data were collected in a structured questionnaire, with Sample/Laboratory ID assigned to each participant and filled with bio-demographic information.

*Sample collection, processing, and storage*

Five (5) mL of blood in plain bottles and urine samples were collected from the same subject in a sterile, labelled container. The sera were separated from the whole blood by centrifugation at 5000 rpm, and the sera were transferred into a labelled sterile cryogenic vial. The urine was filtered, and all samples were stored at -20°C until ready for assay.

*Genomic viral RNA extraction*

Total RNA was extracted from sera and urine using the QIAamp viral RNA mini kit (QIAamp, Germany) according to the manufacturer's instructions. The Laboratory work was conducted at the Nigeria Centre for Disease Control (NCDC) Molecular Laboratory of Aminu Kano Teaching Hospital, Kano, Nigeria.

One-step RT-PCR for cDNA synthesis and PCR amplification in a single reaction was performed by the addition of 2 x Reaction mix-10 µL, template RNA-5 µL, F-primer and R-primer (10 µM- 0.8 µL each, enzyme mix (x20)- 1 µL, and Nuclease-free water- 2.4 µL. All the mixtures were added to the reaction tubes and shaken briefly before running on the real-time PCR machine (MIC tubes). The primers and probes used for the study: Real Time PCR, ZIKV-F-5′-UTR 5' TGAYAAGCARTCAGACAC 3', position 1222-1239; ZIKV-R-5′-UTR 5' TCACCARRCTCCCTTTGC 3', position 1302-1319, and product size 98bp, ZIKV- P-5′-UTR 5' FAM-GTGGAYAGAGGYTGGGGAAA-TAMRA 3' 1265-1284.

The primers and probes used were adapted from Yang *et al.* (2017) and synthesized by Inqaba Biotech West Africa Ltd, Co. The qRT-PCR assay conditions were as follows: Reverse transcription for 10 minutes at 55 °C for 1 cycle and initial denaturation for 1minute at 95 °C for 1 cycle. The denaturation process is performed for 10 seconds at 95 °C, and the extension at 30 seconds at 60 °C for 40 cycles with a cutoff value of 31 ±2.5 for Serum and 32± 2.8 for urine (Figures 1 and 2).

The positives identified were subjected to RT-PCR before sequencing. The PCR kit was produced by New England Biolabs Inc. The positive samples were amplified using RT- PCR technology for sequencing. The primers were adapted from Kurosaki *et al.* (2017) and synthesized by Inqaba Biotecc West Africa Ltd, Co. ZIKV-F, 5′-UTR 5′GGAGTCAGGATGGTACTTGTACC-3′ position, 9205-9227; ZIKV-R 5′-UTR; 5′AAAATTGGATATTCAGGAACC3′, position 9938-9958 and product size of 931bp. The master mix was constituted in a biosafety cabinet. One Taq One-step Reaction mix (2x)- 25 µL, one Taq One-step enzyme mix(25x)-2 µL, then gene-specific forward primer and reverse primer (10 µM)- 2 µL each. RNA 5 µL and nuclease-free water were added to a 50 µL total reaction volume. The Program for the thermal cycler (Applied Biosystems- GeneAmp® PCR system 9700) was created before setting up the reaction. The thermal cycler was pre-heated to 45 °C for cDNA synthesis, followed by PCR amplification. cDNA synthesis in 15 minutes, incubation at 48 °C, and pre-denaturation in 1 minute, all at 1 cycle, incubation at 94 °C. Denaturation in 15 seconds at 94 °C, annealing 30 seconds at 55 °C, extension for 1 minute at 68 °C, all for 40 cycles, and final extension for 5 minutes at 68 °C – 1 cycle. Holding time at 4 °C for ∞ - 1 cycle.

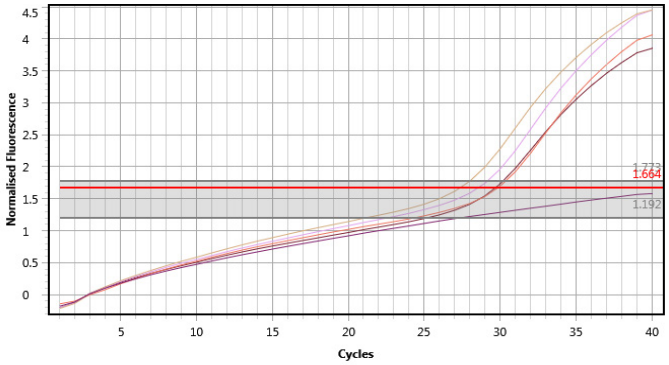


**Figure 1:** *Real Time RT-PCR amplification of viral RNA from Serum samples. Ct Value: 31 ±2.5*

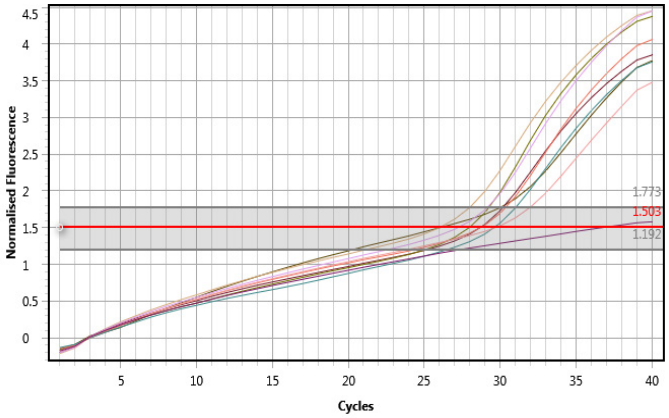


**Figure 2:** *Real-time RT-PCR amplification of the viral RNA from Urine samples Ct Value: 32± 2.8*

*Gel preparation, electrophoresis, and sequencing*

The PCR products were separated using 2% agarose gel. A 100 mL (2%) gel was prepared by dissolving 2 g of agarose powder in 100 mL of 1X TAE buffer. The mixture was gently stirred and microwaved for 3 minutes until fully dissolved and clear. The flask was carefully removed from the microwave, allowed to cool, and then 5 µL of ethidium bromide was added. It was mixed until homogeneous, then poured into the gel casting tray with the comb set properly. After the gel solidified, the comb was removed carefully, and the tray was placed into the electrophoresis tank containing 1X TAE buffer. The DNA ladder, samples, and negative control were loaded. The electrophoresis was run at 140 volts for 40 minutes. Afterward, the gel was transferred to the gel documentation unit and imaged. A photograph of the gel was taken with an in-built camera under UV light (Figure 3). The isolates were sequenced using Sanger sequencing technology. The data collated were analysed using IBM® SPSS® Statistics for Windows (version 27.0.1.0, New York).

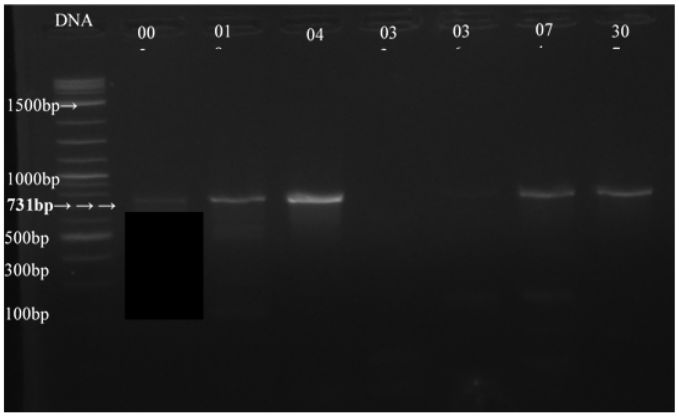


**Figure 3:** *Documentation of gel electrophoresis presentation of some positive samples: Lane 1- 100bp DNA Ladder; Some Positives -Northern Senatorial Zone-( 005, 019, 049, 074), and Southern Zone (307), and Negatives (033, 036).*

## Results

*Socio-demographic and clinical characteristics of the participants*

The socio-demographic information of the respondents in Table 1 and Figure 2 showed that data from 324 respondents were collected, which reveals many characteristics. The mean age of the respondents was 27.10 years with a standard deviation of 5.17. The largest age group was 26-30 years, accounting for 33.6% (n=109) of the respondents, followed by those aged 21-25 years at 27.8% (n=90). Respondents 31-35 years old made up 18.2% (n=59), followed by those 20 years or younger. Women older than 35 years were the least represented 14.8% (n=48). The majority of the respondents were married, representing 94.1% (n=305). Only a few of them were single (3.1%, n=10) or categorized as "others" (separated/single mother) at 2.8% (n=9).

**Table 1:** *Socio-demographic and clinical characteristics of the participants.*

| Variable | Frequency (n= 324) | Percentage (%) |
|---|---|---|
| **Age group (years)** | | |
| ≤20 | 48 | 14.8 |
| 21-25 | 90 | 27.8 |
| 26-30 | 109 | 33.6 |
| 31-35 | 59 | 18.2 |
| >35 | 18 | 5.6 |
| Mean = 27, SD = 10±5.17 | | |
| **Marital status** | | |
| Married | 305 | 94.1 |
| Single | 10 | 3.1 |
| Others (separated/single mother) | 9 | 2.8 |
| **Age of pregnancy (Trimester)** | | |
| First | 56 | 17.3 |
| Second | 164 | 50.6 |
| Third | 104 | 32.1 |
| **Religion** | | |
| Christianity | 259 | 79.9 |
| Islam | 65 | 20.1 |
| **Educational level** | | |
| Non-formal | 48 | 14.8 |
| Primary | 37 | 11.4 |
| Secondary | 147 | 45.4 |
| Tertiary | 92 | 28.4 |
| **Occupation** | | |
| Business | 117 | 36.1 |
| Full time housewife | 87 | 26.9 |
| Student | 40 | 12.3 |
| Farmer | 37 | 11.4 |
| Civil servant | 34 | 10.5 |
| Others | 9 | 2.8 |

Regarding pregnancy trimester, most respondents were in their second trimester (50.6%, n = 164), followed by the third trimester (32.1%, n=104), and then the first trimester (17.3%, n = 56). For religion, Christianity was the predominant faith, accounting for 79.9% (n = 259) of respondents, while Islam represented 20.1% (n=65).

**Table 2:** *Socio-demographic and clinical characteristics of the participants.*

| Variables | Frequency | Percentage (%) |
|---|---|---|
| **Number of children** | | |
| None | 112 | 34.6 |
| One | 91 | 28.1 |
| Two | 80 | 24.7 |
| Three | 27 | 8.3 |
| Above three | 14 | 4.3 |
| **History of miscarriage** | | |
| Yes | 79 | 24.4 |
| No | 245 | 75.6 |
| **Recent history of miscarriage** | | |
| Yes | 32 | 9.9 |
| No | 292 | 90.1 |
| **Recent history of travel outside Nigeria** | | |
| Yes | 5 | 1.5 |
| No | 319 | 98.5 |
| **History of blood transfusion** | | |
| Yes | 12 | 3.7 |
| No | 312 | 96.3 |
| **History of travel outside Nigeria by the husband** | | |
| Yes | 10 | 3.1 |
| No | 314 | 96.9 |
| **Ever lived in a Refugee camp** | | |
| Yes | 15 | 4.6 |
| No | 309 | 95.4 |

Looking at educational attainment, the largest group had a secondary education (45.4%, n=147), followed by those with tertiary education (28.4%, n=92). A notable proportion had non-formal education (14.8%, n= 48), and 11.4% (n= 37) had completed primary education.

Finally, the occupational distribution showed that business owners formed the largest group at 36.1% (n=117). Full-time housewives comprised 26.9% (n=87), while students made up 12.3% (n=40). Farmers accounted for 11.4% (n=37), and civil servants were 10.5% (n= 34). The remaining 2.8% (n= 9) fell under "others" for occupation.

For the number of children, the largest proportion of respondents, 34.6% (n=112), reported having no children, 28.1% (n=91) had one child, while 24.7% (n=80) had two children. A smaller percentage, 8.3% (n=27), had three children, and the smallest group, 4.3% (n=14), reported having above three children.

Regarding the history of miscarriage, the majority of respondents, 75.6% (n=245), reported no history of miscarriage. Conversely, 24.4% (n=79) of the respondents had experienced a miscarriage. Also, a vast majority, 90.1% (n=292), indicated no recent history of miscarriage, and 9.9% (n=32) reported a recent history of miscarriage.

Looking at the recent history of travel outside Nigeria, the data showed an overwhelming majority, 98.5% (n=319), had not recently travelled outside Nigeria, while a very small proportion, 1.5% (n=5), reported recent international travel. Also, for the history of travel outside the country by the husband majority, 96.9% (n=314) of respondents stated that their husband had not travelled outside the country, and 3.1% (n=10) of the respondents reported their husband had a history of international travel.

For the history of blood transfusion, the information showed 96.3% (n=312) reported no history of blood transfusion, while 3.7% (n=12) had a history of blood transfusion.

Lastly, those who had ever lived in a refugee camp showed a substantial majority, 95.4% (n=309), reported never having lived in a refugee camp, while 4.6% (n=15) of the respondents had previously lived in a refugee camp.

### *Prevalence of ZIKV infection among pregnant women in Plateau State, Nigeria*

Zika virus was initially checked from urine samples and sera of the participants by q-RT-PCR. Of all the samples collected from the 324 women, 22 (6.8%) urine samples showed positive test results, and the prevalence was 4.8% (16/324) using sera. This suggests that urine could be more sensitive than serum for the detection of ZIKV using q-RT-PCR (Table 3). Therefore, the overall prevalence was 6.8% (22/324), of which 10 were from the North zone, 7 from the South, and 5 from Plateau Central.

**Table 3:** *Prevalence of ZIKV Infection among Pregnant Women in Plateau State, Nigeria.*

| Sample | Number tested | Number positive (%) |
|---|---|---|
| Serum | 324 | 16 (4.9) |
| Urine | 324 | 22 (6.8) |

**Table 4:** *Genetic diversity of ZIKV among pregnant women in Plateau State, Nigeria.*

| Isolate | Genotype | Lineage (Sub-lineage) | Likely geographic origin |
|---|---|---|---|
| NZ-074 | Asian | American | USA-related lineage |
| NZ-005 | Asian | American | USA related lineage |
| NZ-044 | Asian | American (Mexican Brach) | Mexico Related |
| SZ-257 | Asian | American (Mexican Brach) | Mexico Related |
| NZ-049 | Asian | American-Asian Bridge | Mixed USA-Singapore |
| NZ-024 | Asian | American (Caribbean/USA) | Caribbean and USA linked |
| NZ-009 | Asian | American (Caribbean/USA) | Same lineage as isolates |
| CZ-148 | Asian | American (Caribbean/USA) | Same lineage as isolates |
| NZ-018 | Asian | American (Caribbean/USA) | Same lineage as isolates |

*Key: NZ, Northern Zone, SZ, Southern Zone, Central Zone.*

### *Genetic diversity of ZIKV among pregnant women in Plateau State, Nigeria*

From Table 4, out of the 22 positive samples identified by q-RT-PCR, nine (9) were subjected to RT-PCR and successfully sequenced by the Sanger Sequencing Technique. It can be deduced that, out of the 9 isolates sequenced, seven (7) from the Northern zone, one (1) from the Central Zone, and one (1) from the Southern Zone all belonged to the Asian genotype of ZIKV. Most of them (except for isolate NZ-049, which belongs to the Asian lineage) are within the American lineage. Furthermore, four (4) belong to the American/USA sub-lineage, two (2) to the Mexican sub-lineage; however, two (2) of the isolates were not classified under any sub-lineage. None of the isolates was from the African genotype.

## Discussion

Zika virus infection is still a major worldwide health issue, especially because of the terrible consequences it has on expectant mothers and growing babies. This study offers vital information on the molecular epidemiology and current incidence of ZIKV in pregnant women in Plateau State, North-Central Nigeria.

The sociodemographic information gathered from the individuals showed that the largest age group was 26-30 years, with a mean age of 27.10. Most participants had secondary or tertiary education, were married, and were in the second trimester of pregnancy. These pregnancies are particularly susceptible to ZIKV problems, as seen by the significant percentage who reported having no children or just one. Although the participants' or their husbands' low rates of recent international travel (1.5% and 3.1%, respectively) imply that initial introductions may be travel-related, subsequent transmission is most likely local and may occur through sexual contact or the Aedes mosquito vector (CDC, 2024).

The ZIKV RNA was found to be present in 6.8% (22/324) of urine samples and 4.9% (16/324) of serum samples, indicating continuous transmission in the area even in the absence of a significant epidemic.

This finding is consistent with previous sero-epidemiological data from Nigeria, such as the 6% IgM and 4% IgG positivity reported in North Central Nigeria in 2016 (Carteaux *et al.*, 2016), and the 14.4% overall seroprevalence in Plateau State reported by Anejo-Okopi *et al.* (2020), and 6.1% in a cross-sectional study in Zambia, Babaniyi *et al.* (2015). This is slightly higher than the one reported by Sani et al. (2022), revealing a 4.5% prevalence of Zika IgM antibodies in the sera of participants in a study conducted in Dutse, Jigawa. This is equally higher than the result reported by Adekola *et al.* (2023); in a study conducted in Northwest (Ahmadu Bello University Teaching Hospital, Zaria) and Southwest (Onabanjo Olabisi University Teaching Hospital, Sagamu), Nigeria, who reported an 11.3% prevalence rate with RT-qPCR in serum samples, but 17.2% for anti-Zika IgM among pregnant women. Given the possibility of congenital Zika syndrome, pregnant women should be especially concerned if ZIKV RNA is detected, as this indicates an active infection (Pérez *et al.*, 2025). Urine may be a more sensitive sample type for molecular diagnosis, particularly in the subacute phase of infection, as evidenced by the higher detection rate in urine (6.8% vs. 4.9%) compared to serum, which is consistent with earlier reports indicating prolonged ZIKV shedding in urine (Bingham, 2016; Gourinat *et al.*, 2015). The

results per sample type are also consistent with those of Conceição et al. (2023) in Brazil, who found that urine had a greater positive yield than serum. This also agrees with the study reported by Calvet *et al.* (2019) in Brazil, where urine showed more sensitivity than serum. This has important implications for diagnostic strategies in endemic and outbreak settings.

A striking discovery emerged from the molecular characterisation results: all nine of the isolates that were sequenced were of Asian genotype, and they were mostly grouped under the American lineage, which included the Mexican and American/USA sub-lineages. Additionally, one isolate showed mixed USA-Singapore traits and an American Asian Bridge lineage. Surprisingly, none of the sequenced samples had an African genotype. The historical knowledge of ZIKV circulation in Africa, where the African genotype has historically been endemic since its identification in Uganda in 1947, is significantly different from this (Dick, 1952). The first documented ZIKV infection in Nigeria in 1975 also involved an African strain (Moore *et al.*, 1975). The high incidence of Asian/American genotypes in Plateau State indicates that these strains were introduced and established, most likely as a result of imports associated to travel (CDC, 2024). Travel from other countries is common in Nigeria, and this can help spread arboviruses from other areas (Balint *et al.*, 2021). The sub-lineages that have been observed especially those associated with the United States and Mexico, suggest that they may have ties to areas that have been significantly affected by the current ZIKV outbreak in the Americas. This discovery highlights the worldwide scope of arbovirus transmission and the ongoing necessity of being vigilant against imported strains, which may exhibit distinct epidemiological and clinical features.

In the Americas, severe epidemics and consequences, such as Guillain-Barré syndrome and microcephaly, have been mostly linked to the Asian genotype (Pielnaa *et al.*, 2020; Beber *et al.*, 2023). Consequently, more public health awareness and preventative actions are required due to the prevalence of these genotypes in Plateau State.

It's interesting that the sequenced samples did not include the African genotype ZIKV. Despite the small sequencing sample size (9 out of 22 positive), this study provides insight into the present genetic status of ZIKV in Nigeria. Perhaps imported Asian strains have outcompeted or grown more common, the African genotype circulates at low levels, or the sample technique unintentionally overlooked African strains. To fully map the genetic variation and evolutionary trends of ZIKV throughout Nigeria, larger-scale genomic surveillance studies are required in the future.

For Nigeria and Plateau State, the study's outcomes have significant public health effects. Stronger prenatal screening procedures and public health campaigns aimed at expectant mothers are required due to the occurrence of the Asian genotype, which is linked to serious fetal outcomes. The removal of mosquito breeding grounds and the encouragement of personal protective equipment to prevent mosquito bites are still crucial vector control strategies (Kolawole *et al.*, 2020; Balint *et al.*, 2021). Campaigns to raise awareness of ZIKV sexual transmission should also be stepped up.

## Conclusion

The Zika virus is still circulating among pregnant women in Plateau State, Nigeria, according to this study, with an overall incidence of 6.8% found in urine samples. Importantly, among the sequenced isolates, the molecular characterization showed that the Asian genotype ZIKV was present, primarily from American sub-lineages, while the historically endemic African genotype was not detected. Given their established link to serious neonatal problems, this implies that exotic ZIKV strains were introduced and established, most likely through travel, posing a serious risk to public health.

Based on these findings, the following recommendations are crucial for public health action in Plateau State and across Nigeria:

- Strengthen surveillance system: Implement robust, continuous molecular surveillance programs for ZIKV, particularly focusing on pregnant women and individuals presenting with ZIKV-like symptoms, to promptly detect and characterize circulating strains.
- Capacity building on diagnosis: train laboratory personnel to prioritize the use of urine samples for ZIKV molecular diagnostics due to their potentially higher sensitivity, and ensure laboratories are equipped with the necessary reagents, storage facilities, and expertise for

accurate and timely testing.

- Public health awareness campaign: Launching of a targeted public awareness campaign, especially for pregnant women, regarding ZIKV symptoms, modes of transmission (mosquito bites and sexual contact), and preventive measures.
- Environmental hygiene/vector: Intensify integrated vector control strategies, including source reduction, larviciding, and adult mosquito control, to reduce *Aedes* mosquito populations in residential areas and healthcare facilities.
- Travel health advisories: Provide updated travel health advisories to individuals travelling to and from ZIKV-endemic regions, emphasizing prevention measures.
- Further research: Conduct larger-scale, longitudinal genomic epidemiology studies to comprehensively map the genetic diversity, evolutionary patterns, and transmission dynamics of ZIKV across Nigeria, and to ascertain the relative prevalence of African versus Asian genotypes. Investigate potential links between specific genotypes and clinical outcomes in the Nigerian context.

By implementing these recommendations, public health authorities can better prepare for and respond to ZIKV threats, ultimately safeguarding the health of pregnant women and preventing congenital ZIKV-related complications in Nigeria.

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## Novelty Statement

The research shows that urine is a more sensitive medium than serum for detecting ZIKV RNA using q-RT-PCR, indicating a prevalence of 6.8%. Additionally, this study offers the first molecular evidence that the Asian genotype (notably the American and American Asian Bridge lineages) is circulating among pregnant women in Plateau State, Nigeria.

## Author's Contribution

Conceptualization, ABD, DLR, AIK, and YM; methodology, ABD, DLR and SAD.; software, ABD and DLR.; validation, ABD, DLR, YM, and AIK; formal analysis, ABD, AIK, and DLR; investigation, ABD, SAD and DLR.; resources, ABD and SAD; data curation, ABD, and SAD; writing original draft preparation, ABD, AIK, YM, SAD; writing review and editing, ABD, SAD, DLR, AIK and YM.; visualization, ABD; supervision, DLR, AIK and YM; project administration, ABD and SAD. All authors have read and agreed to the published version of the manuscript.

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*Ethical statement*

We obtained ethical approval for the study from the Plateau State Hospitals Management Board (No. HMB/ADM/772/1/173) and the Plateau State Ministry of Health Research and Ethical Committee (No. MOH/MIS/209/VOL.T/X). We obtained signed or thumbprint-informed consent from all research participants before data collection

*Consent for publication*

All participants provided informed consent.

*Generative AI and AI assisted technology statement*

The authors declare that no generative AI and AI assisted technology was used in the creation of this manuscript.

*Conflict of interest*

The authors have declared no conflict of interest.

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