

Application of lateral flow immunosorbent assay for the detection of Dengue virus antibodies (IgM, IgG, IgM/IgG) among some febrile subjects attending healthcare facilities in Port Harcourt

Abstract

Background: Dengue fever is an infection with similar symptoms as other febrile illnesses such as malaria. It is thus, very important that its possible risk factors and prevalence in areas prone to mosquito infestation be investigated for objective-based focused intervention. This cross-sectional study was carried out using the application of lateral flow immunosorbent assay to determine the seroprevalence of Dengue virus IgM and IgG among febrile subjects in Port Harcourt metropolis.

Method: A total of 110 febrile subjects (already confirmed malaria-positive cases) were recruited from healthcare centres in the study area. Furthermore, a well-structured questionnaire was used to generate their demographics and possible risk factors. The data generated were analyzed using Statistical Package for Social Science (SPSS) version 25. Using a confidence interval of 95%, the *P*-value of <0.05 was considered significant for the analysis.

Results: The prevalence of Dengue virus IgM was 5.4%, IgG 10.2%, and IgG/IgM 4.2%, with an overall prevalence of 9.1%, while Dengue and malaria co-infection was 9.1%. Apart from occupation, there was no significant association between seropositivity, demographics, and risk factors analysed based on Chi-square analysis ($p < 0.05$).

Conclusion: There is therefore, an urgent need for more public health awareness and improvement of surveillance to help mitigate the burden of the disease in our communities.

Keywords: Dengue fever virus, co-infection, IgM and IgG, lateral flow method, risk factors, seroprevalence

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Introduction

Dengue, also known as break-bone fever, is a zoonotic viral infection which is transmitted to humans through the nibble of the female mosquito *Aedes aegypti*.¹ The arboviral disease can also be secondarily spread by the *Aedes albopictus* mosquito. About 2 billion people are at risk of infection by at least one of the four Dengue virus subtypes (DENV-1, DENV-2, DENV-3, and DENV-4).² Recently, a fifth serotype, DENV-5 was discovered. The infection can be in form of Dengue haemorrhagic fever (DHF) or Dengue shock syndrome (DSS) and both types can be fatal.³ The Dengue virus belongs to the genus *Flavivirus* in the family *Flaviviridae*.⁴ These viruses are positive-stranded RNA viruses with a round lipid membrane.⁴ The viral envelope, capsid, and underlying proteins of the layer, as well as seven non-structural proteins are encoded for by the viral genome.⁵

On a global scale, the sharp increase in the prevalence of the virus recorded in recent decades, particularly across world, has caused it to be regarded as a major international public health concern. With an estimated annual incidence of 390 million cases, Dengue virus poses a risk to 2.5–3.6 billion people annually in over 125 endemic countries and has a case fatality rate exceeding 5% in some areas.^{6–8} The virus has become endemic in Asia, Africa, Latin America and the Caribbean, and the Pacific.⁹ Nonetheless, within the past 50 years, Dengue infection has been a public health concern all over the world with a 30% incidence.¹⁰ A report demonstrated a yearly frequency of 390 million cases, of which 96 million (24.6%) were symptomatic and

the larger part were as asymptomatic.⁸ Factors responsible for Dengue virus transmission include population, urbanization, increased global travel, and worldwide environment changes.¹¹

Infection by any of the serotypes may lead to either asymptomatic infection or a febrile illness displaying vary clinical manifestations ranging from acute to more chronic illness e.g., Dengue haemorrhagic fever (DHF) and Dengue shock syndrome (DSS).^{12,13} Acquiring any one of these serotypes will typically confer lifelong immunity to disease caused by that serotype, but not necessarily protection against the other serotypes, so individuals may be affected up to four times over their lifetime.¹⁴ DENV has the potential to cause a range of disease symptoms, from asymptomatic infection (50–75% of infections), to mild, to severe, and even deadly manifestations.

Since the first isolation of DENV in Nigeria, there has been a paucity of reports on this infection, possibly due to the unavailability of sufficient diagnostic tools in health institutions and low awareness by healthcare providers.^{15,16} This is despite reports of the virus being actively circulating in various parts of the country.^{16,17} Many patients with fever are designated as having a fever of unknown origin or malaria or typhoid and remain without a diagnosis even if they fail to respond to antimalarial or anti-typhoid chemotherapies. Under these prevailing practices, there is a real potential of misdiagnosing Dengue fever.^{18,19} The aim of this research was therefore, to determine the sero-prevalence of Dengue fever in Port Harcourt, Nigeria. The impact of misdiagnosis of the disease which is mistaken for other

fever-like illnesses was reviewed using structured questionnaire in order to guide public health intervention program in addressing the menace in malaria prone communities.

Methodology

Study area

The samples were obtained from consented subjects with febrile syndrome visiting healthcare centres and hospitals in Port Harcourt. Port Harcourt is the capital city of Rivers State in Nigeria. This is the centre of oil and gas activity in Nigeria with massive influx of multi-national companies and high human traffic of expatriates and regional citizens who are moving into the city for greener pasture. Nonetheless, the latitude of Port Harcourt is 4.82416, while the longitude is 7.033611. coordinates of Port Harcourt is 4° 49' 27.0012" N and 7° 2' 0.9996" E respectively.

Study design and technique

The study design utilized health centre-and-hospital-based cross-sectional research design. A total of 110 subjects were randomly selected with an inclination to those who presented with fever symptoms. The random sampling technique was used based on the inclusion and exclusion criteria. Utilizing the cross-sectional pilot study design, the subjects were randomly selected especially subjects with febrile conditions.

Sample size calculation

To determine the minimum sample size, the previous seroprevalence of DENV IgM detected in febrile children in South-Western Nigeria of 2.3%, was adopted²⁰ using the formula below

$$N = \frac{z^2 pq}{d^2}$$

Where N = Minimum sample size

Z = Standard normal deviation corresponding to 95% confidence level set at 1.96

p = 2.3% = 0.023, q = 1 - p = 0.977, d = desired precision, 5% (0.05)

$$N = \frac{1.96^2 \times 0.023 \times 0.977}{0.05^2} = 35$$

For improved statistical analysis power, the sample size was increased to 110 for this study to increase research space of inclusivity that promotes more febrile subjects to be included in the study.

Pre-sample collection/advocacy

Samples were collected after awareness and consent was obtained from the Healthcare care services, hospitals, guardians and parents of subjects based on the eligibility criteria. Only those who gave oral consent were pre-selected for the study while those who did not, were not selected for the study. A well-tailored questionnaire was administered to obtain the subject socio-demographics from healthcare centres, hospital records, and also parents and guardians.

Inclusion and exclusion criteria for selecting research subjects

Inclusion criteria were febrile subjects in the study area only, febrile subjects that have given consent for their samples to be used for the study, those with massive pains in their joints, and those already diagnosed of malaria with fever ≥ 38 °C. The exclusion criteria were

non-febrile children, men and women, and febrile subjects that did not give consent for their samples to be used for the study.

Sample collection and preparation procedure

Five millimetres (5 mL) of blood were collected aseptically from each consented subject using a sterile needle and syringe. The whole blood was used to make a thin and thick blood film on a grease-free glass slide (for confirmation of malaria parasite). The serum was separated by centrifugation at 1500 rpm for 5 minutes and stored at -20 °C until it was required for DENV analysis. The serum samples were analyzed for Dengue NS1 and IgM/IgG antibodies using the One Step Dengue NS1 Antigen and IgG/IgM Antibody Duo Panel RapiCard™ InstaTest kits, based on the manufacturer's instruction. The blood film on the glass slide was stained using the Giemsa stain solution and microscopy was carried out to detect and identify the presence of malaria parasites as a confirmation of malaria positive samples and to also increase the quality control mechanism of the study respectively.

Questionnaire

A well-structured questionnaire was administered to all subjects to determine their socio-demographics, as well as the risk factor (s) promoting the spread of the infection.

Ethical consideration

Ethical approval was obtained from the Rivers State Central Ethical Committee on Innovative Research and Development, and written consent was obtained from patients before questionnaire administration and sample collection respectively

Data analysis

The data collected was organized via Microsoft Excel, and subsequently exported into Statistical Package for the Social Sciences (SPSS) version 23 to analyze the data at 0.05 level of significance.

Results

Seroprevalence of anti-dengue virus antibodies (IgG, IgM, IgG/IgM) and malaria co-infection

Figure 1 shows the overall prevalence of 9.1% (10/110) of Dengue virus among the recruited subjects; IgM 4 (5.4%), IgG 6 (10.2%) and IgG/IgM 2 (4.2%).

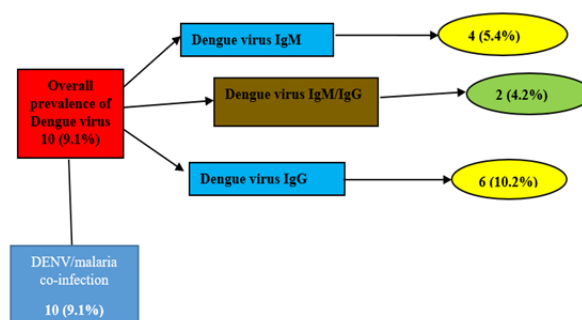


Figure 1 Prevalence of Dengue virus IgM, IgG, and IgG/IgM among febrile subjects.

Distribution of Dengue virus IgM/IgG based on demographics of subjects

Table 1 shows the distribution of Dengue virus IgM/IgG based on the demographics of subjects. Male subjects had a higher DENV

IgG prevalence of 8.1% while female subjects had 2.1%. Based on age, the 16-20 years group had a higher DENV IgG prevalence of 15.4% compared with other age ranges. Single subjects recorded an IgG prevalence of 6.5% and the married subjects had 4.2%. Women

without children had a prevalence of 5.9% while there were no positive cases for those with one child. Those with secondary school education had a prevalence of 2.7% each for IgM and IgG/IgM while unemployed subjects had an IgG prevalence of 5.6%.

Table 1 Distribution of Dengue virus IgM/IgG based on demographics of subjects

		Dengue virus IgG/IgM Frequency			
		Negative	IgG	IgM	IgG/IgM
Gender	Female	43 (89.6%)	1 (2.1%)	2 (4.2%)	2 (4.2%)
	Male	55 (88.7%)	5 (8.1%)	2 (3.2%)	0 (0.0%)
Age range	0-5	6 (85.7%)	1 (14.3%)	0 (0.0%)	0 (0.0%)
	6-10	15 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	11-15	8 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	16-20	11 (84.6%)	2 (15.4%)	0 (0.0%)	0 (0.0%)
	21-25	3 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	26-30	13 (86.7%)	0 (0.0%)	1 (6.7%)	1 (6.7%)
	31-35	1 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	36-40	17 (85.0%)	1 (5.0%)	1 (5.0%)	1 (5.0%)
	40-45	1 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	46-50	13 (86.7%)	1 (6.7%)	1 (6.7%)	0 (0.0%)
	51-55	1 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	56-60	8 (80.0%)	1 (10.0%)	1 (10.0%)	0 (0.0%)
	71-75	1 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Marital status	Single	57 (91.9%)	4 (6.5%)	1 (1.6%)	0 (0.0%)
	Married	41 (85.4%)	2 (4.2%)	3 (6.3%)	2 (4.2%)
Number of children	0	61 (89.7%)	4 (5.9%)	2 (2.9%)	1 (1.5%)
	1	2 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	2	8 (88.9%)	1 (11.1%)	0 (0.0%)	0 (0.0%)
	3	11 (84.6%)	0 (0.0%)	1 (7.7%)	1 (7.7%)
	4	10 (90.9%)	1 (9.1%)	0 (0.0%)	0 (0.0%)
	5	6 (85.7%)	0 (0.0%)	1 (14.3%)	0 (0.0%)
Pregnant	No	98 (89.1%)	6 (5.5%)	4 (3.6%)	2 (1.8%)
Educational level	None	8 (80.0%)	1 (10.0%)	1 (10.0%)	0 (0.0%)
	Primary	18 (94.7%)	0 (0.0%)	1 (5.3%)	0 (0.0%)
	Secondary	32 (86.5%)	3 (8.1%)	1 (2.7%)	1 (2.7%)
	Tertiary	38 (90.5%)	2 (4.8%)	1 (2.4%)	1 (2.4%)
Employed	Yes	34 (89.5%)	2 (5.3%)	1 (2.6%)	1 (2.6%)
	No	64 (88.9%)	4 (5.6%)	3 (4.2%)	1 (1.4%)
Occupation	Student	24 (92.3%)	2 (7.7%)	0 (0.0%)	0 (0.0%)
	civil servant	9 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	Tailor	1 (33.3%)	1 (33.3%)	0 (0.0%)	1 (33.3%)
	None	31 (88.6%)	2 (5.7%)	2 (5.7%)	0 (0.0%)
	Self-employed	20 (83.3%)	1 (4.2%)	2 (8.3%)	1 (4.2%)
	Retired	1 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)

Distribution of Dengue virus IgM/IgG based on risk factors

Table 2 shows the distribution of Dengue virus IgM/IgG based on risk factors. Those who had no knowledge of Dengue virus transmission recorded the highest seroprevalence of anti-Dengue virus antibodies, such as IgG (10.0%), IgM (4.0%), and IgM/IgG (2.0%). Those who used a hospital as their primary healthcare provider had the highest IgG and IgM prevalence with 7.5% and 5.7%, respectively. Those who did not have healthcare centres around them recorded IgG prevalence of 10.0% while those who had recorded 3.3% and 2.2% for

IgM and IgG/IgM, respectively. Based on home environments those who have not used tyres around their homes had an IgG report of 6.5%, and those who had refuse dumped around their homes reported IgM, and IgM/IgG prevalence of 2.5% and 4.5% respectively. Those who had no knowledge of Dengue fever had IgG occurring in 5.7%, IgM in 3.8% and IgM/IgG in 1.9%. Those who did not know that there was a similarity between malaria and Dengue recorded an IgG prevalence of 5.6%, IgM with 3.7% and IgM/IgM with 1.9%. Those who had no knowledge that DENV was transmitted by mosquitoes recorded a prevalence of 5.7% for IgG, 3.8% for IgM and 1.9% for IgG/IgM.

Table 2 Distribution of Dengue virus IgM/IgG frequency based on risk factors

		Dengue fever IgG/IgM Frequency			
		Negative	IgG	IgM	IgG/IgM
Presence of malaria	Yes	95 (88.8%)	6 (5.6%)	4 (3.7%)	2 (1.9%)
	No	3 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Transmission	Yes	56 (93.3%)	1 (1.7%)	2 (3.3%)	1 (1.7%)
	No	42 (84.0%)	5 (10.0%)	2 (4.0%)	1 (2.0%)
Healthcare used	Chemist	40 (93.0%)	1 (2.3%)	1 (2.3%)	1 (2.3%)
	Pharmacy	13 (92.9%)	1 (7.1%)	0 (0.0%)	0 (0.0%)
	Hospital	45 (84.9%)	4 (7.5%)	3 (5.7%)	1 (1.9%)
Frequency of malaria attack	Every month	35 (85.4%)	4 (9.8%)	2 (4.9%)	0 (0.0%)
	Every two months	19 (95.0%)	1 (5.0%)	0 (0.0%)	0 (0.0%)
	Every 3 months	19 (86.4%)	1 (4.5%)	0 (0.0%)	2 (9.1%)
	Above 3 months	25 (92.6%)	0 (0.0%)	2 (7.4%)	0 (0.0%)
Health centre	Yes	81 (90.0%)	4 (4.4%)	3 (3.3%)	2 (2.2%)
	No	17 (85.0%)	2 (10.0%)	1 (5.0%)	0 (0.0%)
Healthcare distance	5 mins	16 (88.9%)	0 (0.0%)	1 (5.6%)	1 (5.6%)
	<30 mins	46 (88.5%)	3 (5.8%)	2 (3.8%)	1 (1.9%)
	>30 mins	1 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	1 hour	20 (87.0%)	2 (8.7%)	1 (4.3%)	0 (0.0%)
	>1 hour	15 (93.8%)	1 (6.3%)	0 (0.0%)	0 (0.0%)
Laboratory visit	Yes	69 (90.8%)	4 (5.3%)	2 (2.6%)	1 (1.3%)
	No	29 (85.3%)	2 (5.9%)	2 (5.9%)	1 (2.9%)
Mosquito nets	Yes	19 (95.0%)	0 (0.0%)	1 (5.0%)	0 (0.0%)
	No	79 (87.8%)	6 (6.7%)	3 (3.3%)	2 (2.2%)
Plantain or bushy environment	Yes	66 (89.2%)	4 (5.4%)	3 (4.1%)	1 (1.4%)
	No	32 (88.9%)	2 (5.6%)	1 (2.8%)	1 (2.8%)
Used tyres	Yes	42 (87.5%)	2 (4.2%)	2 (4.2%)	2 (4.2%)
	No	56 (90.3%)	4 (6.5%)	2 (3.2%)	0 (0.0%)
Fishing port	Yes	35 (89.7%)	2 (5.1%)	1 (2.6%)	1 (2.6%)
	No	63 (88.7%)	4 (5.6%)	3 (4.2%)	1 (1.4%)
Refuse dump	Yes	38 (86.4%)	2 (4.5%)	2 (4.5%)	2 (4.5%)
	No	60 (90.9%)	4 (6.1%)	2 (3.0%)	0 (0.0%)
Dengue fever knowledge	Yes	5 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	No	93 (88.6%)	6 (5.7%)	4 (3.8%)	2 (1.9%)
Malaria/dengue similarity	Yes	2 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	No	96 (88.9%)	6 (5.6%)	4 (3.7%)	2 (1.9%)
Dengue and mosquito	Yes	5 (100.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	No	93 (88.6%)	6 (5.7%)	4 (3.8%)	2 (1.9%)

Based on the Cramer's V analysis, there was a strong association ($V > 0.5$) between risk of infection and age range, number of children, pregnancy, educational level, employed, presence of malaria, healthcare used, health centre, healthcare distance, laboratory visit, mosquito nets, plantain or bushy environment, fishing port, Dengue fever knowledge, Malaria/Dengue similarity, Dengue and mosquito. There was a medium association (V ranges between 0.4-0.5) reported for used tyres. Also, there was a weak association (V ranges between 0.1-0.3) reported for gender, marital status, occupation, transmission, frequency of malaria attack, and refuse dump.

Discussion

This pilot study was specifically designed to determine the seroprevalence of Dengue virus (DENV) IgM/IgG among febrile subjects in Port Harcourt metropolis. The prevalence of DENV was 5.4%, 10.2% and 4.2% for DENV IgM, IgG, and IgG/IgM respectively. The overall prevalence was 10 (9.1%) and Dengue and

malaria co-infection was 9.1% among the 110 recruited subjects in the study area. A similar cross-sectional study by Onyedibe [2018] in Jos and Maiduguri, Nigeria which recruited 529 febrile patients, recorded the prevalence of anti-Dengue IgM and IgG, as well as NS1 by Enzyme-Linked Immunosorbent Assay (ELISA) to be 2.3%, 5.5% and 1.5% respectively.²⁰ The study by Onyedibe [2018] used a more sensitive molecular technique and a larger sample size compared with our study. In another related study conducted in Nigeria. It was reported that the pooled prevalence of DENV IgM, IgG, RNA, NS1 and neutralizing antibodies were 16.8%, 34.7%, 7.7%, 7.7% and 0.7%, respectively.²² This study also reported a lower prevalence than the 6% reported in a study conducted in Cross Rivers State, southern part of Nigeria.⁵ The researchers strongly believed that the differences observed in the result pattern of different studies could be due to certain dynamics in the various study locations, such as unplanned urbanization, population density, human mobility, access to reliable water source, and water storage practice. Moreover, in addition to

the study design, other likely notable factors such as Dengue vectors adaptability to the environment or climate, population's knowledge, attitude and practice towards Dengue might be attributed to the variations in research outcomes at different geographical settings.

Furthermore, in a gender-based study, the serological difference between Dengue virus NS1 antigen (Ag) and IgM antibody (Ab) among 4252 patients via ELISA were evaluated. The study revealed a higher percentage of positive cases for males (26%) than females (19.5%) of which the difference between them were statistically different ($P < 0.0001$).²³ This finding corroborates the findings of this study in which males had a higher IgG prevalence of 8.1% compared with that of females, 2.1%. Similarly, in a cross-sectional study in Bangladesh among 553 laboratory-confirmed and 194 probable dengue cases, out of which two-thirds were male (63.2%), the average age was 27(+11) years.²⁴ These findings suggest dengue virus infection as male-gender-prone. The probable leading reason that could be linked to more males getting infected than females might be based on the fact that males are more mobile than females in the region where this study was conducted. Men move around to provide for their family, food and daily upkeep. They go to farms and hunt in thick forests to provide food for their family. Thus, such routine for family economic survival, tend to predispose men to mosquito bites, which potentially inoculate them with the Dengue viral pathogen. They also stay longer outside in the evenings and nights for village/community meetings, thereby exposing themselves more to mosquito bites, this is unlikely to women and girls who often stay indoors to cook and look after homes and children, alongside taking care of other eventful domestic works that keep the family moving in a stable and more sustainable manner.

In a cross-sectional study carried out in Vietnam, during the recent outbreak of Dengue virus infection few years ago, the researchers' critical finding was that patients of all ages were affected; however, mortality risk was higher among those with underlying conditions and elderly patients.²⁵ Their finding suggests that immunocompetent subjects were least ravaged by the mosquito-borne viral pathogen, Dengue virus. In addition, the finding further corroborates the general scientific belief that in times of community outbreak of infectious disease, those with terminal or underlying terminal infections, backed up with old age in the community would be more impacted, probably due to their weak immunological response and performance, especially impairment of cell-mediated immune resistance mechanism.

Recently, Kotepui et al., in their systematic review estimated the pooled prevalence of Dengue and malaria co-infection among febrile patients using a random-effects model. The study found out that 32% of patients with Dengue and malaria co-infection had severe malaria.²⁶ Therefore, it is possible to deduce that such co-infection of Dengue and malaria might have a synergistic adverse effect on the host, thereby escalating disease outcome.

Symptoms of febrile conditions known by respondents in this study are fever and joint pain. The study by Hasan *et al.* reported that respondents displayed symptoms such as abdominal pain (86.5%) anorexia and/or vomiting (69.6%), and diarrhoea (26.2%) which was more frequent than typical rash and other pain symptoms.²⁴ In a systematic review, it was reported that symptoms of severe DENV infection were bleeding (39.6%), jaundice (19.8%), and shock/hypotension (17.9%), while that of severe malaria were severe bleeding (47.9%), jaundice (32.2%), and impaired consciousness (7.43%).²⁶

The use of insecticide and mosquito nets in the control of Dengue virus vectors has been reported in a number of studies. In a report 49% of study subjects slept under insecticide-treated mosquito nets in the

past week preceding research and 44% of subjects might have taken antimalarial therapies prior to seeking care.⁵ This finding is lower compared to the 19% of the negative cases and 1% of positive cases that admitted to using nets in this study. Nevertheless, the importance of this study cannot be emphasized, hence the need to deploy much-needed differential diagnosis to rule out diagnostic probability would be helpful and highly appreciated towards improving patients' welfare and reducing the patient's long stay in our health care facilities, due to the misery of misdiagnosis and presumptive reportage of false negative or positive results in the diagnosis of malaria cases that share almost similar striking signs and symptoms with Dengue virus infection in a malaria endemic regions. Further studies would be needed in different parts of Rivers State with the use of Real-Time PCR and ELISA methods, so as to drive the gains of the above pilot study home for the good of all and sundry in our society since "health is wealth".

Limitation

Some febrile patients usually do not visit the healthcare facility but use over-the-counter drugs which makes proper diagnosis difficult.

Conclusion

This study reported the prevalence of Dengue virus IgM 5.4%, IgG 10.2% and IgG/IgM 4.2% with an overall prevalence of 9.1% using Dengue virus immunochromatographic assay among the 110 recruited febrile subjects. There was no statistical significance seen from all the various demographic and risk factors variables analyzed, but there was a positive correlation between socio-demographics/risk factors and seropositivity. There is thus, the need to increase public health awareness of Dengue fever due to its similarities with malaria infection to aid directed medical intervention. The healthcare services and local medical laboratories should be manned by well-trained scientists who can handle the new diagnostic methods available to rule out false positive and negative results. Citizens (especially pregnant women, children and the elderly) should be educated on proper preventive measures to help reduce the morbidity of the infection. The habit of self-medication, since every fever symptom feels like malaria, should be discouraged. Well-funded robust epidemiological and entomological surveillance in Rivers State, Nigeria should be made a priority by policymakers to help fight the menace.

What is already known on this topic

1. That the virus is transmitted by the *Aedes aegypti* mosquito which is present in this sampled area;
2. That symptoms of the virus infection are similar to that of malaria infection;
3. That anyone can be infected regardless of age or gender.

What this study adds

1. The misdiagnosis that occurs between Dengue fever and malaria because both are febrile illnesses.
2. The prevalence of Dengue virus among subjects in Port Harcourt, Rivers State.
3. The perceived risk factors among at-risk population.

Authors' contributions

AO and WGN put together the concept and design of the project as well as the definition of intellectual content; WRC and GMU carried out the literature search, clinical studies, experimental studies, and

data acquisition; AO carried out the data analysis, statistical analysis, manuscript preparation, while GMU carried out manuscript editing and manuscript review.

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Conflicts of interest

The authors declares they have no conflicts of interest.

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