Prevalence of Dengue Virus Infection Among Patients with Febrile Complaints attending Murtala Muhammad Specialist Hospital Kano

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Abstract
Dengue virus is a small spherical, enveloped, mosquito-borne single stranded, positive sense, RNA virus belonging to the Flavivridae family, having a cubic symmetry and measures 40-50nm in diameter. It is the most rapidly spreading mosquito-borne viral disease with an estimated incidence of 390 million per year worldwide. The aim of this study is to determine the prevalence of Dengue viral infection among patients with febrile complaint attending Murtala Muhammad Specialist Hospital Kano. A total of 80 febrile patient attending General Outpatient Department were recruited in this study and 7 (8.8%) were found to be positive for Dengue virus NS1 antigen using enzyme linked immunosorbent assay (ELISA) following manufacturer’s instruction (SD-Standard Diagnostics, Inc. Germany). The age range of 10-14 years has the highest percent age of Dengue NS1 antigen and a statistical significance relationship was found between Dengue virus infection and variable of age with a p value of 0.040. This study concludes that Dengue virus was in circulation in the study area and might be one of the causes of acute undifferentiated fever among febrile patients in Kano.

Keywords: Dengue virus, NS1-antigen, febrile patients, Kano

Introduction
The name Dengue was derived from a Swahili word *kidinga-pepo* meaning a sudden seizure by a demon (Ananthanarayan and Panicker, 2003). The earliest known documentation of dengue-like illness was derived in Chinese encyclopedia during 265 AD (Subhash, 2012). In 1780, Rause coined the term “break borne fever” base on description of symptoms reported by patients during Philadelphia epidemics of probably dengue fever (Mohan, 2005). The
possible dengue outbreaks were documented sporadically every 10 to 30 years until world war II, the first epidemics of dengue hemorrhagic fever was described in 1963 in Manila (Luang-Suarkia et al., 2017). In 1979 to 1980 the first outbreak of dengue fever occurred simultaneously in Asia, Africa, and North America (Subhash, 2012). The virus is a small spherical, enveloped, mosquito-borne single positive-stranded RNA virus belonging to the Flaviviridae family, Flavivirus genus, having a cubic symmetry and measures 40-50nm in diameter (ICTV, 2018).

Dengue virus is transmitted through the bite of infected mosquito of the genus Aedes aegypti and other species of the Aedes (Bhatt et al., 2013). It is the most rapidly spreading mosquito-borne viral disease with an estimated incidence of 390 million per year worldwide (Simmons et al., 2013).

The onset of the fever may be sudden or there may be prodromal symptoms of malaise, chills, and headache (WHO, 2018). Pains soon developed, corresponding to peak viral load. The temperature may subside on about the third day and rise again about 5-8 days after onset (Siraj et al., 2017). A rash may appear on the third or fourth day and last for one to five days, although complication and death are rare. Especially in young children, dengue may be mild febrile illness, and dengue hemorrhagic fever or dengue shock syndrome in individuals usually children (Jawetz, 2013).

Diagnosis of this virus should be considered in persons who develop a fever within two weeks of being in the tropics or subtropics (Simmon et al., 2012). The initial change evident on laboratory investigations is a low white blood cell count, which may afterward be followed by low platelets and metabolic acidosis (Ranjit and Kissoon, 2011). There are ongoing programs working on a dengue vaccine to cover all five serotypes (Normile, 2013). The aim of this study was to determine the prevalence of Dengue virus in patients presented with febrile illness in General Outpatient Department of Murtala Muhammad Specialist Hospital Kano.

Area of The Study
The research was carried out at Murtala Muhammad Specialist Hospital that lies between latitude 11° 30’ N and longitude 8° 30’ E within Kano metropolis 1580 feet above sea level. It is situated in the Sahelian geographic region, south of the Sahara. Kano city is in Kano state which is located in North-western Nigeria.

Sample Size Determination
The sample size was calculated and determined using the formula as follows
\[
n = \frac{z^2pq}{d^2}
\]  
(Lwanga and Lameshow, 1991; Naing et al., 2006).
Where:
\(n\) = number of samples
\(z\) = statistic for level of confidence at 95% = 1.96
\(p\) = prevalence (4.8%) = 0.048 (Moses et al., 2016).
\(d\) = allowable error of 5%, (0.05)
\[
n = \frac{1.96^2 \times 0.048 (1 - 0.048)}{0.05^2} = 70
\]

Study Population
A total of 80 Participants with febrile complaints were recruited in this study, and both were grouped into age and gender.
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Sample Collection
Approximately 2 ml of venous blood sample was collected aseptically from each of the patients under consideration and dispensed in to plain containers (Cheesbrough, 2016).

Processing
The sample was allowed to clot, and centrifuged at 3000 rpm for 5 minutes. The serum was transferred into labeled 2 ml plain containers. The samples were analyzed using enzyme linked immunosorbent assay (ELISA) following manufacturer’s instruction (SD-Standard Diagnostics, Inc. Germany).

Test Procedure
Fifty (50ul) of the sample diluent was added to each well including the control wells. Fifty (50ul) of the negative control were added to the first three wells. Fifty (50ul) of the positive control were added to the fourth and fifth wells aseptically. Fifty (50ul) of each serum sample were added to the respective wells. The micro titter plate was then covered with microplate sealer and was mixed well. The microplate was incubated at 37°C for 60 minutes. The wells were then washed with 350ul of the washing solution using automatic washer. Hundred (100ul) of the enzyme conjugate were added to each well. The microtiter plate was then covered with adhesive plate sealer and was mixed properly. It was incubated at 37°C for 60 minutes. The plate was washed 5 times with washing solution using automatic washer. Hundred (100ul) of tetramethyl benzidine (TMB) were added to each well. The plate was incubated for 10 minute at room temperature which results in blue color development. Hundred (100ul) of stop solution were added to each well. The absorbance was read using spectrophotometer at 450nm.

Results
Table 1 shows that, 7 (8.75%) were found to be positive for Dengue virus NS1 antigen using Enzyme Linked Immunosorbent Assay (ELISA).

Table 1: Distribution of Dengue virus among the study group

<table>
<thead>
<tr>
<th>Condition</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>7</td>
<td>8.8</td>
</tr>
<tr>
<td>Negative</td>
<td>73</td>
<td>91.2</td>
</tr>
<tr>
<td>Total</td>
<td>80</td>
<td>100</td>
</tr>
</tbody>
</table>

Five patients (6.2%) in the age group 10-14 were positive for dengue infection, age group 5-9 years has one positive (1.2%) and greater than 50 years also 1 (1.2%) with P value (0.040) as shown in Table 2.

Table 2: Distribution of Dengue virus infection by age group

<table>
<thead>
<tr>
<th>Age Range (years)</th>
<th>Positive</th>
<th>Negative</th>
<th>Percentage Positive (%)</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>5-9</td>
<td>1</td>
<td>8</td>
<td>1.25</td>
<td>9</td>
</tr>
<tr>
<td>10-14</td>
<td>5</td>
<td>13</td>
<td>6.25</td>
<td>18</td>
</tr>
<tr>
<td>15-19</td>
<td>0</td>
<td>13</td>
<td>0.00</td>
<td>13</td>
</tr>
<tr>
<td>20-29</td>
<td>0</td>
<td>16</td>
<td>0.00</td>
<td>16</td>
</tr>
<tr>
<td>30-39</td>
<td>0</td>
<td>12</td>
<td>0.00</td>
<td>12</td>
</tr>
<tr>
<td>40-49</td>
<td>0</td>
<td>6</td>
<td>0.00</td>
<td>6</td>
</tr>
<tr>
<td>&gt;49</td>
<td>1</td>
<td>5</td>
<td>1.25</td>
<td>6</td>
</tr>
<tr>
<td>Total</td>
<td>7</td>
<td>73</td>
<td>8.8</td>
<td>80</td>
</tr>
</tbody>
</table>

P value 0.040

In the current study patients were divided in to gender of which 49 were males (61.25%) with 4 (5%) out of 7 and females 31 (38.75%) with 3 out of 7 (3.8%) as shown in table 3.
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Table 3: Distribution of Dengue Virus Infection Based on Gender

<table>
<thead>
<tr>
<th>Gender</th>
<th>Positive (%)</th>
<th>Negative (%)</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>4 (8.1)</td>
<td>45 (91.8)</td>
<td>49</td>
</tr>
<tr>
<td>Female</td>
<td>3 (9.7)</td>
<td>28 (90.3)</td>
<td>31</td>
</tr>
<tr>
<td>Total</td>
<td>7 (8.8)</td>
<td>73 (91.2)</td>
<td>80</td>
</tr>
</tbody>
</table>

P value (0.815)

The participant ranges from 5-70 years of age were at different level of education fifteen (15) are in primary school, eighteen (18) secondary school, fourteen (14) tertiary institution and remaining 33 are on non formal education. The subject were further classified according to their occupation of which eleven (11) were civil servant, nine (9) business and thirteen (13) house wives and approximately 47 students were recruited in the study as shown in table 4 and both has no knowledge of dengue virus infection and its sign and symptoms.

Table 4: Educational level and occupation of the participants

<table>
<thead>
<tr>
<th>Educational Level and Occupation of Participants</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary</td>
<td>15</td>
<td>18.75</td>
</tr>
<tr>
<td>Secondary</td>
<td>18</td>
<td>22.50</td>
</tr>
<tr>
<td>Tertiary</td>
<td>14</td>
<td>17.50</td>
</tr>
<tr>
<td>Civil Servant</td>
<td>11</td>
<td>13.75</td>
</tr>
<tr>
<td>Business</td>
<td>9</td>
<td>11.20</td>
</tr>
<tr>
<td>House wife</td>
<td>13</td>
<td>16.25</td>
</tr>
<tr>
<td>Total</td>
<td>80</td>
<td>100</td>
</tr>
</tbody>
</table>

Discussion

Dengue is a single stranded RNA virus of 11kb size that usually causes sudden onset of fever with prodromal symptoms of malaise, chills, and headache (Basilio-De-Oliveria et al., 2005; Amarasinge et al., 2011).

Out of the subjects studied, 7 (8.8%) were found to be Dengue NS1 antigen positive using enzyme linked immunosorbent assay (ELISA) technique. Forty nine (61.2%) of the study subjects were males and 31 (38.75%) of the studied subjects were females.

Gender distributions of the positive cases shows a prevalence of 4 (5%) males while 3 (3.8%) were females with p value of 0.815 that is statistically insignificance.

The results from this study are similar with the findings of Nasir et al. (2017) in Abuja who reported NS 1 antigen prevalence of 8.8%. It is lower than the finding reported in Maiduguri by Hamisu et al. (2017) who reported 9.9% and similarly 13.9% reported in a study in Kenya by Vu et al. (2017). These findings could be due to differences in the geographical locations of the study sites and the time of the year the study was carried out. Current finding is higher than prevalence of 4.8% NS 1 antigen reported in Uyo by Moses et al. (2016). The Uyo which is Deltaic savanna area in southern part of the country with heavy and abundant rainfall, these storms are usually convectional in nature due to the regions proximity, to the equatorial belt. Dawurung et al. (2010) reported a prevalence of 2.2% in Jos by and 0.5% reported in another study in Maiduguri by Baba and Talle, (2011). The lower prevalence reported in the previous studies could be as a result that the vector is not much common in their study areas at that time compared to what is currently obtainable.
However DENV IgM antibody prevalence rate of 17.2% was reported in Ogbomoso, Oyo State by Oladipo et al. (2014), in another study in Ogbomosho, Nigeria, 30.8% IgM antibody were reported by Faneye et al. (2013), 35% IgM was reported in Ibadan by Oyero and Ayukekong (2014), 37.4% were reported in Maiduguri by Hamisu et al. (2017) and Similarly in a study conducted by targeting IgM a prevalence of 51.09% in females against 48.9% in males in Kaduna State, Nigeria where reported by Bello et al. (2014). These higher findings may not be unrelated to the difference in the target parameter and the approach toward detecting the causal agent.

The increase in local and international travel to the dengue endemic region coupled with inherent behavior of not taking sanitary measures toward preventing the development and spread of the vector *Aedes aegypti* may be the reasons for the higher findings. Higher prevalence of Dengue virus infection in this study was found in children between the age of 10-14 years with 5 (6.2%). Current findings are similar to that of Mishra et al. (2018) and Piedrahita et al. (2018) who reported higher seropositivity in age group 10-14 years in their studies but it contrasts the finding of Tsai et al. (2018) who reported an increase seroprevalence of Dengue virus in older age group. Our finding showed that age group of 5-9 years has 1 (1.2%) and greater than 50 years age group also 1 (1.2%). Age was shown to be statistically significant effect with respect to Dengue virus infection in the current study (p= value = 0.040). This is suggesting that the individuals in this age group 10-14 years were actively involved in day time activity and less concerned with the mosquito bite since the vector of the dengue virus (*Aedes aegypti*) is a day time feeder (WHO, 2017). This increases their chance of becoming infected and due to the fact that their immune system is still developing as such not robust to resist infection with dengue virus upon bite with the infected vector.

The vector for the Dengue virus *Aedes aegypti* may appears to be seasonal vector, although can be found throughout the year due to the presence of stagnant water bodies coupled with poor drainage and waste disposal systems conditions that favors the Aedes spp. of mosquito to breeds in this area (Stewart-Ibarra et al., 2013) but more abundant during the raining season because the mosquitoes like to breads in open gutter, stored water in the container for long period, extended rainfall and ambient relative humidity and temperature may favor the breading of the *Aedes aegypti* and other Aedine mosquitoes. Population growth and increase individual movement, urbanization, and the limited financial and human resource are attributed to the emergence of the disease (Guzman and Kouri, 2004).

### Conclusion

This study concludes that Dengue virus infection was one of the causes of acute undifferentiated fever among febrile patients in Kano. Children aged less than 10-14 years were found to be having higher Dengue virus infection and had a greater risk of becoming infected. However, there was no gender vulnerability among the study participants.

### Recommendations

Government should initiate Dengue virus surveillance and commence an integrated vector control programme and provide resources at hospital laboratories in Nigeria to facilitate early diagnosis and management of dengue patients. Physicians should consider the possibility of dengue cases when examining febrile patients.
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References


