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Original Research Article

Seroprevalence and changing trend of dengue in a tertiary care hospital, Bhubaneswar, Odisha: Four-year retrospective study

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ABSTRACT

Background: Dengue virus is a single stranded positive sense RNA virus belonging to the genus flavivirus family flaviviridae. Dengue fever is a seasonal and emerging acute mosquito borne arbo-viral illness affecting tropical and sub-tropical countries. This illness ranges from a mild asymptomatic form to severe dengue hemorrhagic fever (DHF) with or without dengue shock syndrome (DSS).

Aim: This study was conducted to know the seroprevalence and changing trend of Dengue virus in a tertiary care hospital, Bhubaneswar, Odisha, India.

Materials and Methods: Over a period of four years from August 2016 to December 2019, a total of 5147 blood samples from clinically suspected dengue patients were received in department of Microbiology laboratory. Serum was separated and subjected to enzyme immunoassay for detection of both Non Structural (NS1) antigen and IgM antibody.

Results: During this study period, a total of 5147 blood samples were processed from suspected dengue cases, out of which 1314(25%) samples were found to be positive by different serological markers like NS1 Antigen (Ag), IgM antibody(Ab), or both NS1 Ag & IgM Ab. The overall seroprevalence rate was found to be 25%. In this study period of four years, the year-wise seroprevalence rate was found to be 12% (153) in the year 2016 and was 26% (350) in the year 2017 and was 40% (522) in the year 2018, and was 22% (289) in the year 2019. It clearly shows that there is an increase in the dengue cases.

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1. Introduction

Dengue virus is a single-stranded positive-sense RNA virus belonging to the genus *Flavivirus* and family *Flaviviridae*. Dengue fever is a seasonal and emerging acute mosquito-borne arboviral illness affecting tropical and subtropical countries. This illness ranges from a mild asymptomatic form to severe dengue hemorrhagic fever (DHF) with or without dengue shock syndrome (DSS). The globally estimated burden of symptomatic cases ranges to 58.4-96 million cases/year.¹ In 2009-2017 in India, 683,545

dengue cases and 2,576 dengue deaths were reported from surveillance data as per the national vector-borne disease control program.² The case fatality rate due to dengue is >1 over the last decade.³ Dengue virus has five different serotypes, indicating that immunity is serotype-specific.⁴ All the five serotypes are prevalent, causing epidemics in India now and then. The virus is transmitted from humans to humans through the vector, the *Aedes aegypti* mosquitoes, and sporadically by *Aedes albopictus*.⁵ Each serotype of the virus produces specific, lifelong immunity but only short-term cross-immunity.⁶ The estimated mortality rate is 2.5%, and the endemicity is spread across 128 countries. However, epidemic outbreaks are more common during the

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rainy season, when the vector population is higher.⁷ The two factors associated with the increased severity of dengue infection are secondary dengue infection and infection with a virulent viral strain.⁸ Early diagnosis of dengue infection remains the cornerstone for treatment and prevention of dreadful complications such as DHF and DSS.⁹

For any virus infection, the standard serological test, hemagglutination inhibition, neutralization test, indirect immunofluorescence antibody test, enzyme-linked immunosorbent assay (ELISA), complement fixation test, or rapid immunochromatography test can be used. Out of these tests, ELISA is the most widely used method for routine diagnosis of dengue infection for its high sensitivity, specificity, and its simplicity and cost-effectiveness.¹⁰ Detection [Editor 9] of non-structural, highly conserved glycoprotein-1 (NS1) antigen is a novel approach for the diagnosis of acute dengue, as it was found to be circulating in the blood during the acute phase of the disease from the first to the ninth day of fever.^{11,12}

With this background, this 4-year retrospective study aimed to describe the frequency, distribution and changing trend of dengue infection in Bhubaneswar, Odisha, India.

2. Materials and Methods

This study was a retrospective observational study conducted in the department of Microbiology at a tertiary care hospital in Bhubaneswar after obtaining Institutional Ethics Committee approval (Ref. No.: KIIT/ KIMS/ IEC / 343/2020). The data were collected from medical records at the Department of Microbiology of the hospital for a period of 4 years and four months from August 2016 to December 2019. The study included patients who were clinically suspected of dengue. The World Health Organization criteria for the diagnosis of dengue was followed.

Universal safety precautions were followed while collecting and processing blood samples from patients. Blood samples (3 mL) taken from patients under clinical suspicion of dengue viral infection with a short history and duration of fever on the day of presentation in the hospital was submitted to the Microbiology Department. A total of 5,147 non-repetitive blood samples were collected during the study period and sent to the microbiology laboratory to test for dengue virus infection. Hemolyzed and lipemic samples were excluded from the study.

Blood samples were stored in the refrigerator at 4°C-8°C and processed for serum separation within 24 h. Serum was separated by centrifuging blood at 3,000 rpm for 5 min. The separated serum samples were subjected to serological testing, depending on the duration of fever at the time of presentation of the patient to the hospital (less than/more than five days). Samples were respectively chosen to be processed for NS1 antigen detection and IgM antibody detection.

NS1Ag and IgM antibodies were detected using Dengue NS1Ag capture ELISA and IgM capture ELISA (Pan Bio Dengue diagnostics Abbott). The positive control [Editor 4] and Negative Control from the test kit were put up. The ELISA microtiter plates were read with a Bio-rad ELISA reader. Optical density values were recorded and analyzed, and the results were read according to the manufacturer's instructions.

Relevant sociodemographic and clinical data of patients with dengue were obtained from medical records and analyzed.

3. Result

During this study period (August 2016 to December 2019), a total of 5147 blood samples were processed from suspected dengue cases, out of which 1314(25%) samples were found to be positive by different serological markers like NS1 Antigen(Ag), IgM Antibody(Ab), or both NS1 Ag & IgM Ab. The overall seroprevalence rate year wise was found to be 25%, as shown in Table 1.

Table 1: Year-wise distribution of seroprevalence rate of dengue positive cases

Year	Total number of suspected dengue cases	Number of positive dengue cases
Aug. 2016-Dec.2016	490	153 (12%)
2017	1172	350 (26%)
2018	1863	522 (40%)
2019	1622	289 (22%)
Total	5147	1314(25%)

3.1. The trend of dengue infection

In this study period of four years, the year-wise seroprevalence rate was reported to be 12% (153) in the year 2016 and was 26% (350) in the year 2017 and was 40% (522) in the year 2018, and was 22% (289) in the year 2019 as shown in Table 1. The trend of dengue infection over the four years period is represented in Table 2 and Figure 1. It clearly shows that there is an increase in the dengue cases after June and July, and also, there is the highest peak in the cases following the rainy seasons (September-November). During the January to May months, there is a drop in the reported cases.

Out of 1314 positive dengue cases, the seroprevalence of NS1 (Ag) was 28% (375), IgM (Ab) was 43% (559), and both NS1 and IgM Ab positive was 29% (380) as shown in Table 3 and Figure 2.

In total dengue positive cases (1314), affected males were 69% (909), and females were 31% (405). So males are 2.2 times more affected as compared to females as shown in Table 4.

Table 2: Seasonal variation of dengue/ month wise distribution of clinically diagnosed and serologically positive cases from 2016-2019

Month	2016 (n)	2017 (n)	2018 (n)	2019 (n)
January		2	9	1
February		4	13	1
March		4	24	0
April		1	23	7
May		1	12	9
June		6	56	13
July		16	42	24
August	35	77	90	42
September	47	46	123	53
October	44	92	84	64
November	20	80	33	54
December	07	21	13	21
Total=1314	153	350	522	289

Table 3: Distribution of different serological markers (NS1Ag, IgM Ab, or both NS1 Ag +IgM Ab) of dengue in positive dengue cases

Year	Number of dengue +ve with Only NS1Ag	Number of dengue +ve with Only IgM Ab	Number of dengue +ve with both NS1Ag+IgMAb
2016	38	66	49
2017	73	187	90
2018	197	176	149
2019	67	130	92
Total = 1314	375 (28%)	559 (43%)	380 (29%)

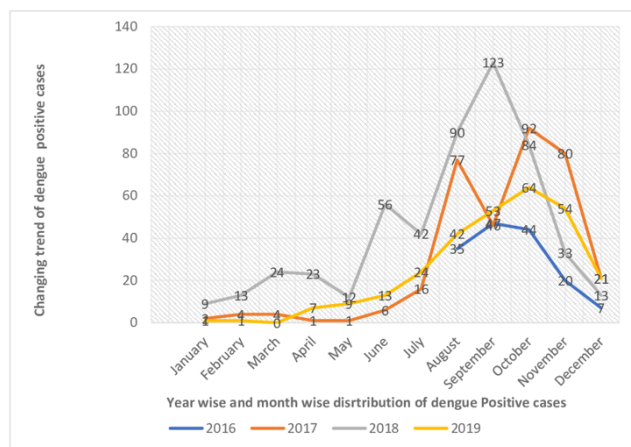


Fig. 1:

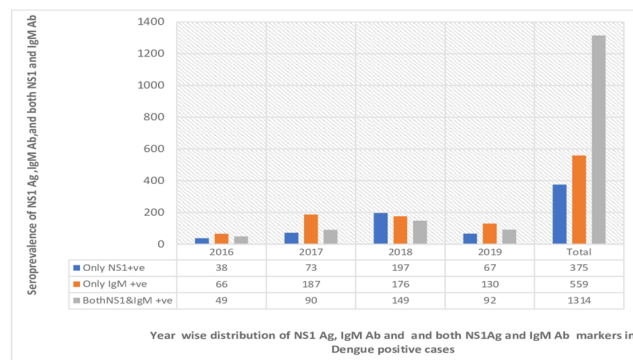


Fig. 2:

Table 4: Gender wise distribution of dengue positive cases from 2016-2019

Year	Male (n)	Female (n)
2016	107	46
2017	237	113
2018	349	173
2019	216	73
Total N=1314	909(69%)	405(31%)

4. Discussion

Dengue fever is an acute febrile viral infection, which has become a significant public health problem in tropical and subtropical regions of the world. In India, the first epidemic of clinical dengue-like illness was recorded in Madras (Chennai) in 1780, and the first virologically proven epidemic of dengue fever occurred in Calcutta (Kolkata) in 1963-1964, wherein 200 people died of it.^{13,14} The first major outbreak of dengue fever/DHF occurred in Delhi in 1996, where 10,252 cases were recorded, and 423 deaths were reported.¹⁵ Dengue is an urban disease, but it has changed character over time. Increased travel among people to neighbouring states for jobs and business might be responsible for the rapid spread of disease to new areas.

Furthermore, unplanned urbanization and poor sanitation facilities create fertile breeding grounds for mosquitoes. Laboratory diagnosis of dengue infection is crucial, as the varied presentation of the disease can make accurate clinical diagnosis difficult. Assays based on the detection of NS1Ag or IgM Ab are commonly used in most the laboratories.

25% of patients had serologically confirmed dengue infection in the present study. A similar surveillance study done by Sood³ reported 18.99%, whereas Garg et al.¹⁶ reported the seroprevalence of dengue infection in their study area to be 19.7%.

For early diagnosis of dengue, our laboratory uses ELISA for NS1Ag and IgM Abs detection. Testing for these two factors would increase the rate of detection of dengue fever at an early stage. A study by Neralwar et al.¹⁷ also showed similar findings. The present study showed that the post-monsoon season (August-November) is the peak season for dengue cases to occur. This should be considered to create a preventive strategy to minimize dengue infection. Testing for NS1Ag and IgM could significantly improve diagnostic sensitivity, which helps in the timely management of dengue infections. Further studies should be done to determine the prevalence of serotypes and genotypes in this area to prevent impending outbreaks due to DHF.

We found that men were more affected than women, constituting nearly two-thirds of the total cases. This was confirmed by Antony et al.¹⁸ (60.70%), Garg et al.¹⁶ (67%), and Kumar et al.¹⁹ (64.6%), although Padhi et al.¹⁰ reported a female preponderance in the cases, they analyzed. [Editor 3] This trend could be because males are more likely to travel and work than females in India. This finding indirectly indicates the importance of workplaces and travel on dengue incidence, which needs further exploration.

An increase in dengue cases was observed after June and July and peaked following the rainy seasons (September-November). This indicates an increase in the breeding places for the vectors during the post-monsoon period.

The limitation of the present study was that we were unable to perform serotyping to determine the prevalent serotypes due to economic constraints and limited resources.

5. Conclusion

Regular epidemiological studies are necessary to monitor the dengue situation in an area, help early detection of an outbreak, and initiate effective control measures. The study results indicate the need for the proper education of the public through various available media and awareness campaigns. Most cases were reported during the post-monsoon period, which warrants coordinated action toward vector control measures. Active participation from the public is an essential component to curb down the problem. There is also an urgent need to develop a vaccine that is

effective against all five dengue virus serotypes.

6. Source of Funding

None.

7. Conflict of Interest


The authors declare no conflict of interest.

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