Section: Microbiology



# **Original Research Article**

# SEROPREVALENCE OF DENGUE IN PATIENTS ATTENDING TERTIARY CARE HOSPITAL, NIZAMABAD, A CROSS-SECTIONAL STUDY

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# ABSTRACT

**Background:** Dengue virus belongs to the genus Flavivirus and family Flaviviridae. Its nucleus consists of single–stranded positive sense RNA. Dengue fever is an acute mosquito-borne arboviral illness affecting sub-tropical and tropical countries. It is a seasonal and emerging disease. Dengue illness is caused by four serologically related viruses designated as DENV-1 to 4 and ranges from mild symptomatic form to severe dengue haemorrhagic fever (DHF) with or without dengue shock syndrome. **Aim & Objectives:** Seroprevalence and evaluation of sensitivity and specificity of the immunochromatographic method to that of the ELISA diagnostic method in the detection of dengue fever at Government General Hospital, Nizamabad.

Material and Methods: Over a period of one year from March 2021 to Feb 2022, a total of 2084 blood samples from clinically suspected cases of dengue were received in the Department of Microbiology laboratory. The serum was separated and subjected to enzyme immunoassay for detection of both Non-Structural (NS1) antigen and IgM antibodies. Rapid dengue method and NS1&IgM ELISA tests were done for the suspected fever cases.

**Results:** During this study period, a total of 2084 blood samples were processed from suspected dengue cases, out of which 381 (18.22 %) 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 18.22 %. Prevalence of NS1 antigen 113 (77%) (n=145). The sensitivity and specificity of the immunochromatographic method were found to be 93 % and 100 % as compared to the ELISA method.

Conclusion: The present study detected NS1 antigen along with IgM Antibodies. The result revealed that both NS1 and IgM have a very high specificity, the sensitivity and specificity of the Immunochromatic method were found to the 93% and 100% respectively as compared to ELISA method. Serological diagnosis should be done in all clinically suspected dengue cases for early initiation of treatment and thereby minimizing mortality. A continuous Sero-epidemiological surveillance control program is needed to overcome future epidemics of dengue, thereby minimizing the complications, outbreaks, and mortality.

Keywords: Non-structural (NS) 1 antigen, Arbovirus, ELISA, IgM antibody.

## INTRODUCTION

Dengue virus belongs to the genus Flavivirus, the family Flaviviridae, its nucleus consists of single–stranded positive sense RNA viruses.<sup>[1]</sup> Dengue fever

is an acute mosquito-bornearbo viral illness affecting sub-tropical and tropical countries, it is a seasonal and emerging disease. <sup>[2]</sup> Dengue illness is caused by four serologically related viruses designate das DENV-1 to 4 and ranges from mild symptomatic form to severe

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dengue haemorrhagic fever (DHF) with or without dengue shock syndrome.[3-4] According to WHO, most cases are under-reported due to asymptomatic dengue illness.<sup>[5]</sup> In 2019, WHO enlisted dengue fever as one of the ten threats to global health.[6] In 2010 there were 2.2 million cases reported and it increased to over 4.2 million in 2019 there are different methods of laboratory diagnosis for dengue viral infection other than clinical findings, which are a must for confirmation of disease. [7-8] There are antigen and antibody detection by serology (Rapid, ELSIA), genome detection (PCR), and viral isolation (no longer used for routine testing). for identification of acute infection, especially the detection of antigens and antibodies are done in the laboratory. [9-10] The most commonly used test in the laboratory for the detection of antigens is NS1 - and the antibody is IgM. IgM to IgG ratio is used to detect primary and secondary dengue infection in recent laboratory diagnosis. [6] NS1 antigen detection helps in the diagnosis of dengue virus in the early course of infection.[11] ELSIA is a well-established diagnostic test highly sensitive but time-consuming and rapid tests could cut down the time required. [12-13]

In the present study, the serum samples were collected from the clinically suspected Dengue patients, and subjected to serological diagnosis (NS-1, IgM). This study, aimed to determine the sensitivity and specificity of the RAPID test to that of ELISA in Determining NS1 antigen and IgM antibodies.

# MATERIAL AND METHODS

This is a cross-sectional study over a period of 12 months March 2021 to February 2022conducted in Government General Hospital, Nizamabad. 5 ml of blood sample was collected under strict aseptic precaution by venipuncture in a plain red vacutainer tube. Centrifuged sample at 3000 rpm for 5 minutes, serum sample separated processed for rapid test (ICT) and ELISA test.

#### **Inclusion Criteria**

- 1. Serum sample collected from a clinically suspected patient with fever (>38.50 C) associated with headache, Retro-orbital pain, myalgia, Arthralgia, and Rash.
- 2. Both outpatients and inpatients are included.
- 3. Patients belonging to both genders and all age groups are included.
- 4. Patients who are willing to participate and give consent are included in the study.

#### **Exclusion Criteria**

- 1. Patients who did not present with symptoms and signs of dengue fever.
- 2. Patients who are not willing to give consent.

#### **Ethical Clearance**

Written consent to participate in the study was obtained from the subjects or their guardians after the full explanation of the study was provided to them. This study was reviewed and approved by the Institutional Ethical Committee, government medical

college & General Hospital, Nizamabad. All data were handled confidentially and anonymously.

# **Method of Collection of Data**

Data entry will be done using M.S. Excel and descriptive and inferential statistics using Statistical Package for Social Sciences (SPSS Version 21).

## **RESULTS**

During this study period, a total of 2084 blood samples were processed from suspected dengue cases, out of which 381 (18.22 %) 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 18.22 %. NS1 antigen prevalence and duration of Illness most seen on day 3 seen in this study 113 (77%) (n=145). IgM antibodies are the first immunoglobulin to appear and are detectable in 50% of patients by days 3-5 after the onset of illness, increasing to 80% by day 5 and 99% by day 10. The sensitivity and specificity of immunochromatographic method were found to be 93 % and 100 % when compared to the ELISA method.

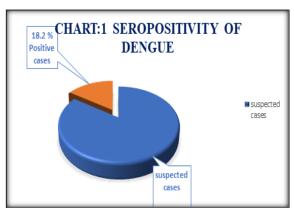


Figure 1: Seropositivity of Dengue

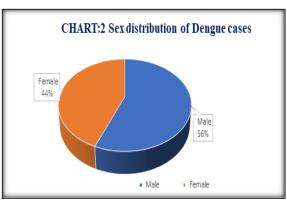


Figure 2: Sex distribution of Dengue cases

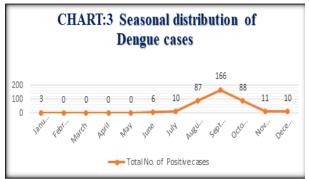


Figure 3: Seasonal distribution of Dengue cases

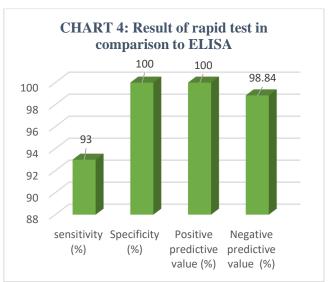


Figure 4: Result of rapid test in comparison to ELISA

Table 1: Prevalence of dengue cases

| Total number of samples tested | Total number of positive samples | % Positive samples |  |
|--------------------------------|----------------------------------|--------------------|--|
| 2084                           | 381                              | 18.2 %             |  |

Table 2: Sex distribution of Dengue cases

| Sex    | Total cases | Total cases Positive cases |      |
|--------|-------------|----------------------------|------|
| Male   | 1130        | 214                        | 56%  |
| Female | 951         | 167                        | 44%  |
| Total  | 2081        | 381                        | 100% |

**Table 3: Seasonal distribution of Dengue cases** 

| Months    | Positive cases | Percentage |  |
|-----------|----------------|------------|--|
| January   | 3              | 0.78%      |  |
| February  | 0              | 0          |  |
| March     | 0              | 0          |  |
| April     | 0              | 0          |  |
| May       | 0              | 0          |  |
| June      | 6              | 1.57%      |  |
| July      | 10             | 2.67%      |  |
| August    | 87             | 22.83%     |  |
| September | 166            | 43.56%     |  |
| October   | 88             | 23.09%     |  |
| November  | 11             | 2.88%      |  |
| December  | 10             | 2.62%      |  |

Table 4: Result of rapid test in comparison to ELISA

| True<br>positiv |   | True<br>negative | False-<br>negative | sensitivity (%) | Specificity (%) | Positive<br>predictive<br>value (%) | Negative<br>predictive<br>value (%) |
|-----------------|---|------------------|--------------------|-----------------|-----------------|-------------------------------------|-------------------------------------|
| 357             | 0 | 2060             | 24                 | 93              | 100             | 100                                 | 98.84                               |

#### **DISCUSSION**

The incidence of Dengue varies in different parts of the country. In this study, we report 18.22% seropositivity, which is like the study conducted by Trupti Bajpaiet.al & Nissi Mathew et.al which showed 18.4% & 19.4% seropositivity respectively. Depending on various factors the South Asia regions are divided into three categories. India in category "A" based on the severity of Dengue is the leading cause of hospitalization and deaths in children in INDIA is hyperendemic with all 4 serotypes in circulation. The prevalence of dengue and its

associated mortality has significantly increased in our country over the last few years. Here in the present study, we report complete demographic and serological studies during the years 2021 to 2022. Over the last few decades, dengue has emerged as a major public health problem in the Indian subcontinent. The rapid increase in the global dengue burden and the inability to accurately diagnose clinically because of its wide clinical spectrum, ranging from mild febrile illness to severe syndrome has promoted our interest in using laboratory diagnosis of dengue infection. In the present study Rapid dengue method, IgM ELISA, and IgG ELISA

were used for 2084 samples (Table 8). Rapid NS1 test was positive for samples 132 (35%), Rapid IgM test was positive for samples 225 (59%), NS1 ELISA in 146 samples (38%), and IgM ELISA in 235 samples (62%). The detection of dengue cases was more by the ELISA method when compared to the Rapid methods in this study. In the study by Man meet Kaur Gillet al., in 2016, they also found that the detection of dengue was more by ELISA than Rapid Method.

# **CONCLUSION**

Dengue fever causes significant morbidity and mortality in developing countries like India and present like any viral illness. Hence these patients should be diagnosed early and treated. The present study showed that NS1 antigen detection along with IgM tests by rapid and ELISA methods. The result revealed that both NS1 and IgM have a very high specificity. The Prevalence of dengue was 18.22 % (381) Among patients attending the fever clinic and patients admitted with fever in Government General Hospital, Nizamabad. A preponderance of dengue was found in males (56%) as compared to females (44%). Dengue has been a major public health problem in India. Based on the present crosssectional study, moderate cases throughout the year and reaching peaks during the post-monsoon period have proved their endemicity and seasonal variation in our region. Community awareness, early diagnosis and management, and vector control measures need to be strengthened to reduce the increasing number of dengue cases. The present study's common age group of patients presenting with dengue was 0 - 20 years. It draws attention toward the male and paediatric, young adult population. NS1 antigen prevalence and duration of Illness most seen on day 3 seen in this study 113 (77%) (n=145). Cases Detection of the dengue NS1 antigen during the symptomatic phase of illness represents an important advance in the diagnosis of dengue fever and is useful for the detection of dengue viral infections early in the course of the infection, especially in non-endemic countries antibodies are the first immunoglobulin to appear. These antibodies are detectable in 50% of patients by days 3-5 after the onset of illness, increasing to 80% by day 5 and 99% by day 10. Our finding suggests that the NS1 antigen-capture ELISA is a very useful and specific tool for the diagnosis of acute dengue infection. However, the sensitivity of the NS1 assay depends on the level of viremia and the host humoral immune response. It is abundant in the serum of patients during the early stage of infection. The present study showed that NS1 antigen detection along with IgM tests. The result revealed that both NS1 and IgM have a very high specificity. Serological diagnosis should be done in all clinically

suspected dengue cases for early initiation of treatment and thereby minimizing mortality. A continuous Sero-epidemiological surveillance control program is needed to overcome future epidemics of dengue, thereby minimizing the complications, outbreaks, and mortality.

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