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Original Research Article

Seroprevalence of dengue virus infection by detection of NS1 antigen

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ABSTRACT

Dengue is the most common arboviral infection prevalent in India. Aedes aegypti mosquito is the principal vector followed by Aedes albopictus.It presents in three clinical stages. It is diagnosed by detection of NS1 antigen, IgM antibody by ELISA and viral RNA by reverse transcriptase PCR. Early diagnosis and treatment is essential to prevent the complications of Dengue. The study was done in a Government Thiruvannamalai Medical College & Hospital about the seroprevalence from the month July 2021 to December 2021 by detection of NS1 antigen. The seroprevalence was found to be 11.4%.The highest number of patients (38.2%) belong to the age group between 21-40 years. Highest number of positive (39.5%) cases reported in October month.

1. Introduction

Dengue virus (DENV) is the most common arboviral infection prevalent in India belongs to Flaviviridae. It has four serotypes (DEN-1 to DEN-4). Recently, the fifth serotype (DEN-5) was discovered in 2013 from Bangkok. Aedes aegypti mosquito is the principal vector followed by Aedes albopictus. They bite during day time. A.aegypti is a nervous feeder. Dengue divides into three clinical stages Dengue fever (DF), Dengue hemorrhagic fever (DHF) and Dengue shock syndrome (DSS). In 2009 WHO Classification of Dengue based on severity of infection such as Dengue with or without warning signs and severe dengue. Outcome of dengue depends upon infecting serotype, sequence of infection and age. Dengue is endemic in more than 100 countries with 2.5 billion people at risk. About 50 million of Dengue cases occur every year worldwide, out of which five lakh cases proceed to DHF. Last decade every year more than 1,00,000 cases of dengue with > 200 deaths occur in India.[1] In 2019 >1.37 lakh cases were reported with >130 deaths. All four dengue serotypes have been isolated from India. Serotypes prevalence varies between seasons and places, but DEN-1 and DEN-2 are widespread.[1] It is diagnosed by detection of NS1 antigen, IgM antibody by ELISA and viral RNA by reverse transcriptase PCR. Early detection of the Infection by NS1 antigen by ELISA and Rapid diagnostic test (RDT). RDT for Dengue IgM antibodies or NS1 antigen are available but have poor sensitivity and specificity. Government of India has passed an order in 2016, Rapid diagnostic test considered as probable diagnosis, must be confirmed by ELISA.[2–4]

2. Materials and Methods

This is cross sectional study done in Department of Microbiology, Government Thiruvannamalai Medical College & Hospital between July 2021 to December 2021, and six months period. The blood samples were collected under aseptic precautions. Serum were separated after centrifuging for 2 minutes at 1000 rpm and that serum were tested by ELISA for NS1 Antigen by J Mithra kit.

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2.1. Inclusion criteria

Any age group of patients who were attended outpatient, inpatient departments and emergency services with presenting complaints of less than seven days of fever, headache, rash, retroorbital pain, myalgia and other symptoms suspicious of Dengue fever.

2.2. Exclusion criteria

Fever confirmed by known infectious causes such as Typhoid and Hepatitis and other non-infectious causes.

2.3. Sample collection and processing

About 3-5 ml of blood was collected from patient under aseptic precautions. Then serum were separated after centrifuging the blood at 1000 rpm for 2 minutes and were subjected to ELISA testing for NS1 antigen by the JMitra kit (Microwell ELISA Test).

2.4. Procedure

Dengue NS1 Ag MICROLISA is a screening test designed for the qualitative detection of Dengue NS1 antigen in serum or plasma. The kit detects DEN1, DEN2, DEN3 and DEN4 subtypes of dengue. Principle is a enzyme linked immunosorbent assay (ELISA) based on the "Direct Sandwich". The microtitre plate wells are coated with Anti-dengue NS1antibodies, samples which were added in the microwells followed by addition of enzyme conjugate (monoclonal anti-dengue NS1antibodies linked to Horseradish Peroxidase enzyme (HRPO)). A sandwich complex was formed in the well. Dengue NS1 antigen from patient's serum was sandwiched between the antibody and antibody HRPO conjugate. The unbound conjugate was washed off during washing with wash buffer. In addition of the substrate buffer and chromogen, a blue color develops. The intensity of blue colour was proportional to the dengue NS1 antigen concentration in sample. To limit the enzyme-substrate reaction, stop solution was added and a yellow colour develops which read at 450nm spectrophotometrically. The kit sensitivity was found to be 99.5% for Dengue NS1 Ag Microlisa kit.

3. Results and Discussion

The total 710 patients serum were tested in Microbiology laboratory for the period of six months from July 2021 to December 2021. In these 710 samples out of which 387 (54.5%) were males and females were 323 (45.5%).(Figure 1)

The seropositivity for Dengue NS1 by ELISA were 81 (11.4%) out of which 45 (55.5%) were males and 36 (44.4%) were females (Figure 2).

Dengue has become common in tropical and sub-tropical countries. Epidemics are frequently seen in different parts

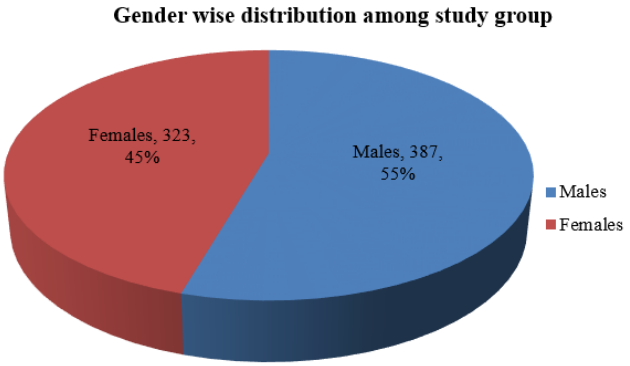

Fig. 1: Gender wise Distribution among the study group

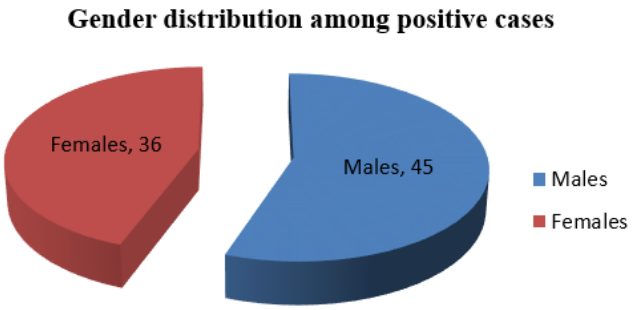

Fig. 2: Gender wise distribution among positive cases

of India especially after the onset of the rainy season. There is no prevention in the form of any vaccine for dengue, thus early diagnosis and treatment is recommended for preventing complications and disease control in the endemic regions. The total 710 serum samples were tested in Microbiology laboratory for the period of six months. 710 out of which 387(54.5%) were males and 323 (45.5%) were females. Dengue seropositivity in our study showed 11.4% out of which males were 45 (55.5%) and females were 36 (44.4%).

Gopal et al., (2016) and Gupta et al., reported a seroprevalence 50% and 29.09% respectively, Kalaivani et al., (2016) reported a 62% of seroprevalence which is very higher compared to our study.[4–6]

The Figure 2 Our study also supports an increased prevalence towards the male gender than female, but reported by Kalaivani et al., (2016) shows equal prevalence. Males are mainly involved in outdoor activities compared to females.

Madan et al., (2018) and Sujatha et al., (2016) reported maximum prevalence in the age group of 21-30 years of age group 26.47% and 31.58% respectively. Mahesh Kumar et al., (2015) reported high prevalence in the age group 10-20 years. Bhat et al., (2013) reported an increased prevalence in adults > 15 years. In our study (Figure 3) showed high

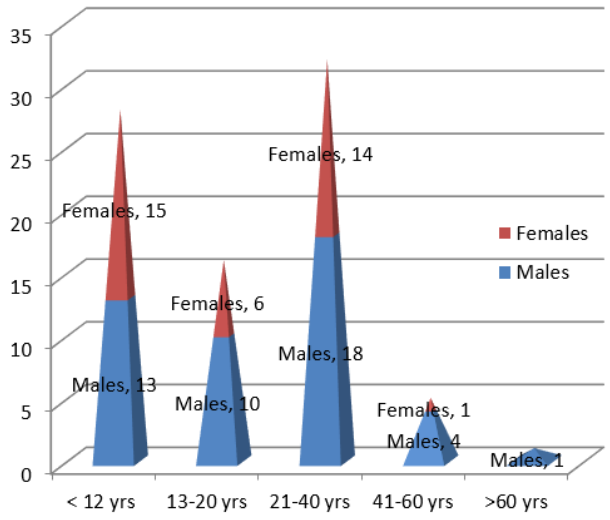

Fig. 3: Dengue positivity distributed among agewise

prevalence of seropositivity recorded in the age group of 21-40 years was 38.2% followed by 34.5% in the age group of <12 years.[7–9]

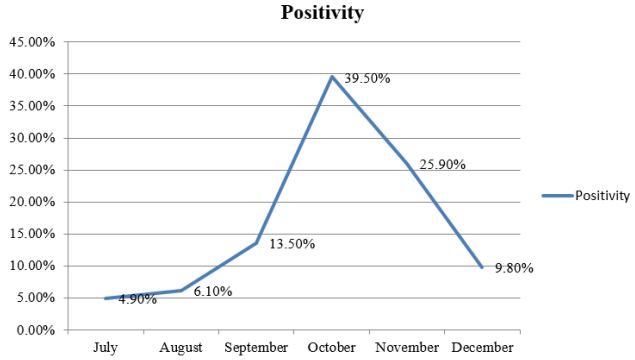

Fig. 4: Represents the prevalence of dengue in relation to months

The highest number of cases were recorded in October and November according to Sujatha et al., (2016). Mahesh Kumar et al., (2015) recorded maximum cases in November followed by October and December. Bhat et al., (2013) also recorded highest number of cases in the season after rainfall.

Patankar et al., (2014) reported increased positivity in October as like as, our study also reported increased positivity in October (39.5%). But Nissi Mathew et al., (2017) study recorded high prevalence during August which is different from other studies. Our study also highlighted the fact that the prevalence of dengue infectivity rate is high in the period after monsoon.[10,11]

Vector control is the best preventive measure by anti-adult measure such as residual spraying, space application and individual protection. In anti-larval measures are larvicide, source reduction and biological larvicide.

Vaccine development for dengue has been a challenge as it should be effective against all four serotypes. After so many trials, recently a vaccine has been licensed for human use since 2015 commercially available dengvaxia. In this vaccine live attenuated yellow fever 17D virus as vaccine vector in which the target genes of all four dengue serotypes are integrated by recombinant technique. It is indicated 9-45 years of age. Schedule: 3 injections of 0.5 ml administrated subcutaneously at 6 month intervals. Currently the vaccine is approved in Mexico, Philippines, Brasil, Indonesia, Thailand, Singapore. In India, it is not available yet because of safety issues.

4. Source of Funding

None.

5. Conflicts of interest

There are no conflicts of interest.

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