European Journal of Molecular & Clinical Medicine

ISSN 2515-8260 Volume 09, Issue 07, 2022

ORIGINAL RESEARCH

Comparative Analysis of Ns1 Antigen and IGM Antibody by ELISA of Clinically Suspected Dengue Fever Cases in a Tertiary Care Centre

T. Mounica[1], D. Lavanya[2], N. Padma Priya[3]

[1] 3rd Year Post graduate, Department of Microbiology, Government Medical College, Ongole, AP, India

[2]Assistant Professor, Department of Microbiology, Government Medical College, Ongole, AP, India

[3]Associate Professor & HOD, Department of Microbiology, Government Medical College, Ongole, AP, India

ABSTRACT

Background: Dengue virus is the most common Arbovirus in India. It is transmitted by female mosquito -Aedes aegypti. It is a major public health problem world-wide, especially in tropical and sub-tropical areas. Approximately 100-400 million dengue cases occur every year according to World Health Organization (WHO). Hence early diagnosis of dengue is essential to prevent complications and mortality.

Material and Methods: A total of 94 cases with clinically suspicious of dengue fever were included. Blood samples from the OP/IP patients with <9 days fever were collected under aseptic conditions. Serum was separated by centrifugation and subjected to detection of dengue NS1 antigen and IgM antibodies by ELISA.

Results: Out of 94 samples, 23 were positive for dengue infection. Out of 23, NS1 antigen was present in 7(30.4%) samples, IgM antibody was present in 4 (17.4%) samples, and both NS1 & IgM antibody were present in 12 (52.2%) samples. The sensitivity and specificity of NS1 antigen - 87.5%&82.5%, for IgM - 80%&79.7%, for NS1 antigen + IgM antibody - 92.3% and 87.6% respectively.

Conclusion: NS1 ELISA test method is an effective method for early detection of dengue fever. Early detection will help in treatment at appropriate time to prevent complications. However, the combination of NS1 Ag test and IgM Ab test by ELISA detected high sensitivity and specificity which provides strong evidence for detection of clinically suspected dengue infection.

Keywords: Dengue, NS1 antigen ELISA, IgM antibody ELISA.

Corresponding Author: Dr. T. Mounica, 3rd year Postgraduate, Department of Microbiology, Government Medical College, Ongole, AP, India.

INTRODUCTION

Dengue virus is the most common Arbovirus in India. It is a mosquito borne infection, transmitted by bite of female mosquito- Aedes aegypti. Mostly it is prevalent in tropical and sub-tropical areas where approximately of about 100-400 million dengue cases occur every year according to World Health Organization (WHO). Dengue virus has four different serotypes (DEN-1 to DEN-4) and recently, the fifth serotype (DEN-5) was discovered in 2013 from Bangkok.[1] The infection may be asymptomatic or may lead to classical dengue fever (DF), or dengue hemorrhagic fever (DHF) with or without shock.[5] As the symptoms of dengue virus infection are not enough for clinical differentiation, definitive diagnosis mainly relies on laboratory tests. A rapid and accurate diagnosis of dengue in the acute phase of

illness is important for initiation of therapy and also for early enhancement of epidemic control measures especially in low endemic areas. The diagnosis of dengue virus infection is mainly based on ELISA detecting either NS1 antigen or IgM antibody. Patients show high levels of non-structural protein – 1 (NS1) antigen in their serum after the onset of infection till 1-7 days. Whereas IgM antibodies are produced after 3-4 days of infection, IgM rises rapidly and are usually identified after 5-6 days. It reaches peak at about 14 days and then declines to undetectable levels over 2-3 months. Therefore the present study was undertaken with the aim to detect NS1 antigen and IgM antibody by ELISA and also to compare analysis of NS1 antigen and IgM antibody detection by ELISA for diagnosis of dengue among the clinically suspected cases.

MATERIALS & METHODS

Study design and site: A study was performed at the Department of Microbiology, Government Medical College, Ongole, and Andhra Pradesh, India. The consent was taken from each patient during this study.

Inclusion criteria

- Suspected cases of dengue including all age groups and both sexes.
- Patients with high grade fever, muscle and joint pains and maculopapular rash over chest and upper limbs.

Exclusion criteria

- Patients diagnosed as malaria, chikungunya or typhoid.

Study period, sample collection and processing

A total of 94 samples of clinically suspected dengue cases were collected from MAY 2022 to JULY 2022. Blood samples (3-4ml) were collected under all aseptic precautions. Serum was separated by centrifugation for serological test.

NS1antigen ELISA

Non-structural (NS1) antigen ELISA was performed using Bio rad- Platelia ^{TM} Dengue NS1 Ag kit as per manufacturer protocol.

Dengue IgM antibody ELISA

Dengue antibody detection was done by NIV DENGUE IgM Capture ELISA kit as per manufacturer instructions.

RESULTS

Of the total 94 clinically suspected dengue cases, 23 serum samples were positive for dengue infection .7 serum samples were positive for Dengue NS1 antigen ELISA , 4 for Dengue IgM ELISA and 12 for both NS1 Antigen + IgM Antibody .

Table 1: Comparison of NS1 antigen ELISA and IgM antibody ELISA

Number of samples tested	Total positives	Only NS1 Ag positives	Only IgM Ab positives	Both NS1 Ag + IgM Ab positives
94	23 (24.4%)	7(30.4%)	4 (17.4%)	12 (52.2%)

Among 23 positive cases, 15 (65.2%) were males and 8 (34.8%) were females.

Table 2: Distribution of dengue by gender

Gender	Total cases	Dengue positive cases			Total positive percentage
		NS1 Ag	IgM Ab	NS1 Ag + IgM Ab	
Male	63	4	3	8	65.2%
Female	31	3	1	4	34.8%

[Table 2]: Show gender wise distribution of dengue positive cases. It shows prevalence of dengue infection is more in male (65.2%) than female (34.8%). Similarly the sensitivity and specificity were analysed, for NS1 Ag - 87.5% & 82.5%, for IgM - 80% & 79.7% and for NS1 antigen + IgM antibody - 92.3% & 87.6% respectively.

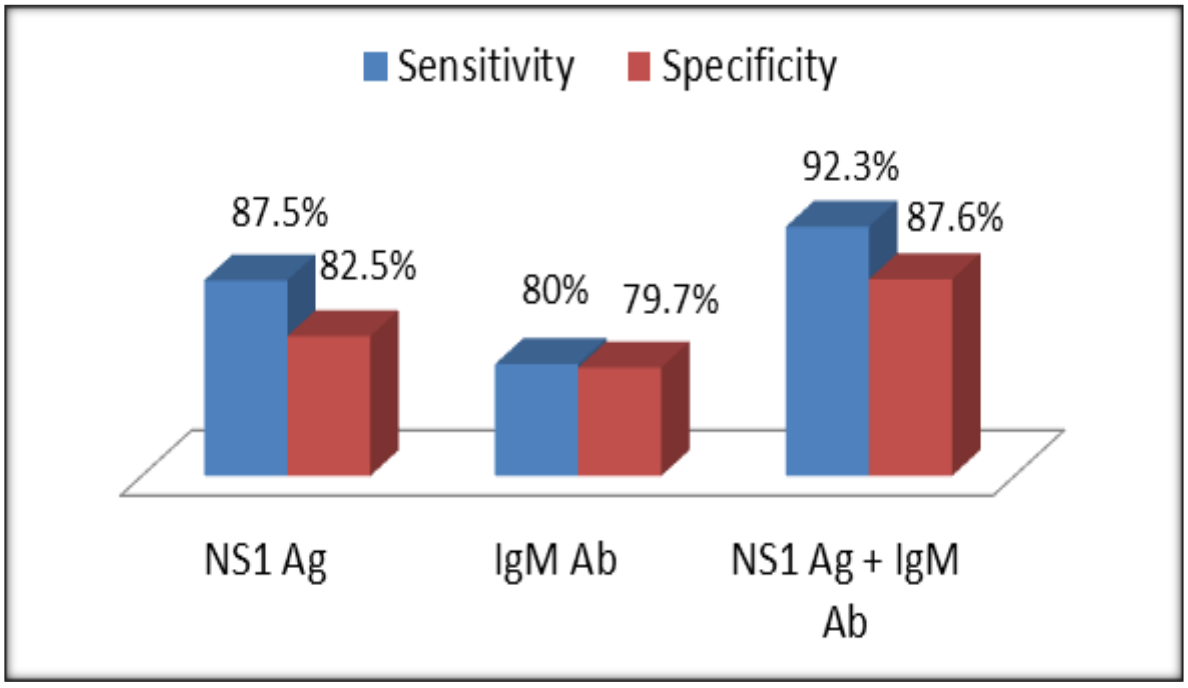

Figure 1: Sensitivity & Specificity

DISCUSSION

Dengue fever is an emerging disease which has high morbidity and mortality. The diagnosis is still great challenge due to lack of resources, infrastructure and skilled manpower in developing countries. This disease is a major health problem in India and needs to be diagnosed and treated in early phase of the disease to prevent complications and also to avoid any associated morbidity and mortality. The effective and accurate diagnosis of dengue is mainly important for clinical care and management. The virus can be detected after onset of illness in serum, plasma, circulating blood cells and other tissues for 1-15 days. Worldwide the serological methods like NS1 and IgM ELISA are routinely being used. In this study also we used NS1 and IgM ELISA as the main diagnostic tool. In this study, the positivity percentage of NS1 Antigen, IgM Antibody and NS1 Ag + IgM Ab was found to be 30.4%, 17.4% and 52.2% which was similar to the study conducted by Dhariti et al.[2] Out of 23 positives cases, 15 were males and 8 females. Thus male preponderance was seen which was similar to the study conducted by Kinal shah et al,[3] and Prakash et al,[7] reported 82.7% total positive percentage in males. The sensitivity and specificity of NS1 antigen - 87.5% & 82.5%, for IgM - 80% & 79.7%, for NS1 antigen + IgM antibody - 92.3% and 87.6%

respectively in our study. Deepak et al, observed sensitivity and specificity of NS1 Ag – 83.58% & 94.82%, for NS1 antigen + IgM antibody -95.55% & 79.31%.[4] The sensitivity and specificity of IgM antibody is similar to the study of Dharitri et al.[2]

CONCLUSION

NS1 ELISA test method is an effective method for early detection of dengue fever. Early detection will help in treatment at appropriate time to prevent complications. As the NS1 antigen is present in serum till 1-7 days and IgM antibodies are produced after 3-4 days. The combination of NS1 Ag test and IgM Ab test by ELISA detected high sensitivity and specificity which provides strong evidence for detection of clinically suspected dengue infection which helps in symptomatic management and reducing morbidity and mortality.

REFERENCES

1. Sastry AS, Sandhya Bhat K, Essentials of Medical Microbiology; second edition 2019;508-511.
2. MAHAPATRA D, Sarangi Gi, ahapatraahapatra Ao, PATY P, DAS P, hayahayani Nu.NS1 Antigen Capture ELISA an Effective Method for Diagnosis of Early Dengue Infection - Report of an Outbreak at Angul District, Odisha, India.J Clin of Diagn Res.2014; 8(8):DC08-DC10.
3. Shah K, Patel V, Shah N.Seroprevalence of dengue infection in clinically suspectedcases of dengue at tertiary care hospital in Gujarat. Int J Med Microbiol Trop Dis 2019;5(3):
4. Kumar D, Verma RK, Singh A, Kumar M, Singh DP, Pandey R, et al. Evaluation of NS1, IgM ELISA and RT-PCR in diagnosis of dengue fever. Int J Res Med Sci 2018;6:2440-3.
5. Neeraja M, V Lakshmi, VD Teja, P Umabala, MV Subbalakshmi. Serodiagnosis of dengue virus infection in patients presenting to a tertiary care hospital. Indian J Med Microbiol. 2006;24:280-82.
6. Dussart P, Labeau B, Lagathu G, Louis P, Nunes MRT, Rodrigues SG, et al.Evaluation of an Enzyme immunoassay for detection of Dengue virus NS1Antigen in human serum. Clin Vaccine Immunology. 2006;13:1185–89.
7. Prakash O, Singh DD, Mishra G, Prakash S, Singh A, Gupta S, et al. Observation on dengue cases from a virus diagnostic laboratory of a tertiary care hospital in north India. Indian J Med Res. 2015;142:7-11.
8. Ngasang AS, Anantapreecha S, Nuegoonpipat AA, Chanama S, Wibulwattanakij S, Pattanakul K, et al. Specific IgM and IgG responses in primary and secondary dengue virus infections determined by enzyme-linked immunosorbent assay. Epidemiol Infect. 2006;134:820-25.