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ORIGINAL RESEARCH

Evaluation of Dengue Rapid Testing and Dengue NS1 & IgM ELISA in Dengue Like Illness Patients Attending at Fever Clinic

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ABSTRACT

Background: Dengue diagnosis is routinely carried out by detection of dengue virus (DENV) antigen NS1 or anti-DENV IgM antibodies using enzyme-linked immunosorbent assays and rapid diagnostic tests. This study was aimed at evaluation of quality of diagnostic assays currently in use in India for the identification of DENV infection.

Methods and Materials: Serum samples from patients (n=200) seeking dengue diagnosis were test educing J Mitra dengue NS1antigen Microlisa kit and J. mitraNS1rapid antigen diagnostic kits. The presence of NS1 antigen was taken as evidence for dengue-positive diagnosis.

Results: In this record review J Mitra ELISA identified 91% patients as dengue NS1positive. J Mitra ELISA taken as reference, the J. mitra NS1 test kit was found to be less sensitive (86.6%) and less specific (89.5%).

Conclusions: These results confirmed superiority of ELISAs for NS1antigen dengue diagnosis, and emphasized on improvement in sensitivity of RDTs.

Keywords: Dengue Virus, Enzyme-Linked Immunosorbent Assay, NS1, Rapid Diagnostic Test

INTRODUCTION

Dengue is one of the most common arboviral viral disease, which is now becoming a major public health concern in India. Dengue virus is positive sense ssRNA belongs to Flaviviridae family. Dengue virus mainly classified into four serotypes (DENV1, DEN-2, DEN-3, DENV-4). Dengue is transmitted by the bite of infected Aedes aegypti and Aedes albopictus mosquito. As per the data of NVPDCP in Gujarat - total 18219 dengue cases were reported in 2019, 1564 in 2020 and 10983 dengue cases in 2021and 4811 dengue cases in 2022 were reported.[1]

Dengue fever is characterized by mild asymptomatic to headache, myalgia/arthralgia, nausea, vomiting and maculo-papular rashes retro-orbital pain, thrombocytopenia which may lead to

fatal dengue hemorrhagic fever (DHF) and dengue shock syndrome(DSS).[2] Other infections like malaria, typhoid, and leptospirosis can mimic dengue and laboratory investigations are essential for an early definite clinical diagnosis.[3-6]

Early detection is important for appropriate treatment and surveillance of disease for proper management and clinical care, particularly for complicated cases and reducing the expenses of investigations and treatments that's why this is a need of hour to establish most sensitive, specific & rapid diagnostic tool for the detection of dengue fever. Dengue can be detected by various techniques which include, dengue rapid test, ELISA, RTPCR, PRNT & Viral culture. In this present study dengue detection was done by rapid detection of Dengue nonstructural 1 antigen and IgM with Dengue NS1 & IgM ELISA.

MATERIALS & METHODS

This study was carried out at Maa Kamla General Hospital - College of Dental Sciences & Research Centre, Ahmedabad under the supervision of department of Microbiology NIMS Medical College, NIMS University Jaipur from July 2021 to dec 2022. Total 200 dengue like illness patients were included in this study. All the patients who were having a fever of less than two weeks were included. 3ml whole blood was collected by venipuncture into a plain vial tube. After 15 mins samples were centrifuged at 3500 RPM for 10 mins for serum separation. Separated serum was used for Dengue rapid test NS1 & IgM by immunochromatography test and Dengue NS1 & IgM ELISA by J. mitra Pvt Ltd.

STUDY DESIGN

Prospective Study

STUDY CENTRE

Department of Microbiology, NIMS Medical College, NIMS University Jaipur Dengue NS1 antigen test was used the human serum by immunochromatography for qualitative detection NS1 antigen of dengue virus. The membrane strip of the device was pre-coated with anti-dengue NS1 monoclonal antibody on test region (T) and goat anti- mouse IgG is pre-coated on control region (C). Positive result was shown a dark pink/purple band on test region followed by band line on control region too.

In this Dengue NS1& IgM ELISA kit, controls and unknown serum samples were diluted in sample dilution buffer containing secondary antibody and incubated in micro titration wells as per the standard protocol. ThepresenceofNS1antigen and IgM antibody on this ELISA kit is confirmed by the colorimetric response obtained using an HRP Conjugate and substrate. All the results were analyzed by SPSS software ver. 21.

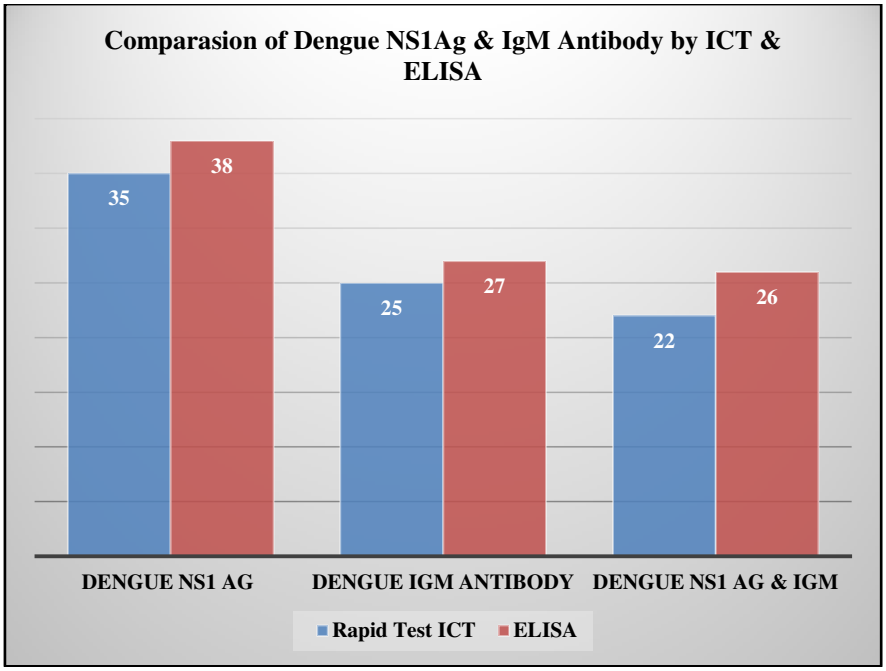
RESULTS

In this present study, during study period total 200 dengue suspected sample were tested for the presence of NS1 antigen & dengue IgM antibody. Out of which total 66 cases were found positive by Rapid card while 91 were found positive by ELISA Method. In this present study male 62.63%positive while Females were 37.26%. The mean age group was 35-45 yrs.

Total 35 cases were found positive for dengue NS1 antigen by Rapid Card while 38 were found positive by Dengue NS1Ag ELISA.

Total 25 cases were positive by IgM Rapid card while 27 were Positive on Dengue IgM ELISA.

Total 26 cases were found positive both NS1Ag and IgM by Rapid card while 26 cases were positive for both NS1Ag & IgM by ELISA.



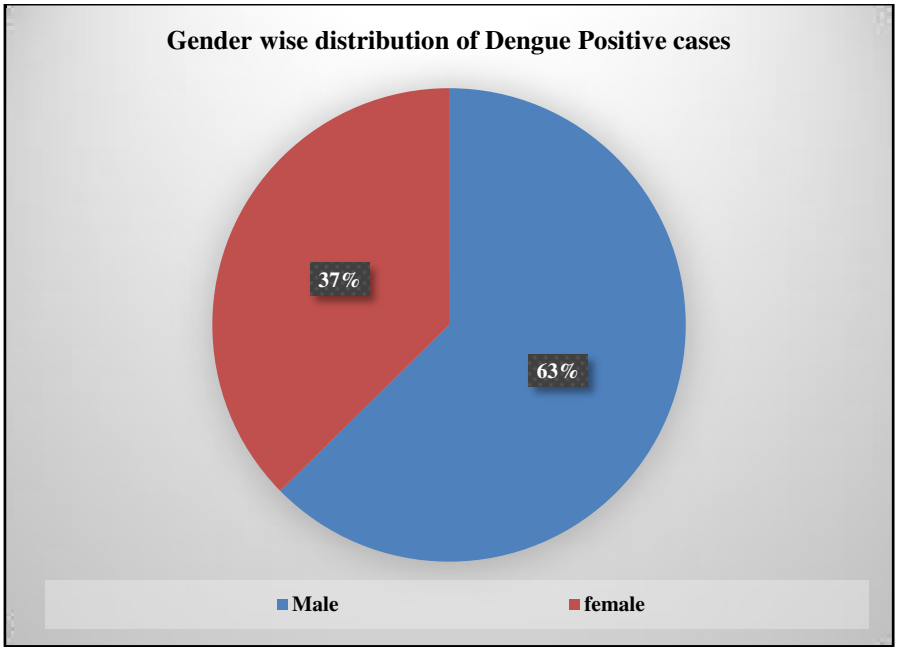


Table 1: Comparative table of sensitivity and specificity of various rapid diagnostic test kits in different studies

Kit used	Serological target	Sensitivity	Specificity	Place of study	Reference number
Bio line Dengue Duo	NS1Ag	70%	73%	Bangkok	17
	NS1Ag	58%	-	Mexico	18
	NS1Ag	76%	98%	Malaysia	19
	NS1Ag	81%	98%	Singapore	20
Dengue NS1	NS1Ag	99%	96%	India	30

DISCUSSION AND CONCLUSION

There is no preventive medication nor any vaccine for dengue, Early diagnosis and treatment

is the key for preventing complications and for disease control in the prevalent region. Proper diagnosis of dengue infection is also difficult because of its symptoms arenot specific, especially in the early stage. The diagnosis of dengue infection can be done through viral isolation, viral RNA detection by RT-PCR, but this method is time consuming and not available in most of the tertiary care hospitals, so diagnosis is based on the detection of dengue specific NS1antigenvia rapid kits or ELISA.

The sensitivity of the rapid ICT tests for NS1 Ag in our study was more than 90%, and specificity more than 95% when compared to ELISA. Hence, the probability of patient suffering from acute dengue infection if the tests are positive is more in ELISA based tests compared to Rapid ICT. Findings of this study are similar with other studies, which have shown the results of rapid ICTs (Pal et al., 2014[13]; Groen et al., 2000[14]; Shih et.al.,2016[15]). Few Indian and foreign studies with similar background are tabulated in Table2. The rapid ICTs have a advantage that they are easy to perform, need less expertise and are completed within minutes (Chaterji et al.,2011[16]) For making ELISA cost effective, large number of samples need to be processed at same time. For ELISA test, lab needs to be equipped with instruments like ELISA washer and reader. Comparison to ELISA, rapid ICT need very less technical expertise to perform and the time for the results is within minutes. The main advantage of the rapid ICT is that a single sample can be run without waiting for the samples.

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