Original Research Article

Comparison of NS1 Antigen detection by Rapid Immunochromatography Test and ELISA for early diagnosis of Dengue at a Government General Hospital, Ananthapuramu

Running Title: COMPARISION OF DENGUE NS1 ANTIGEN WITH RDT & ELISA

ABSTRACT:

Background: the incidence of Dengue hemorrhagic fever, Dengue shock syndrome associated with Dengue can be reduced by diagnosing Dengue early and by initiating early treatment to Dengue patients. This study was conducted to compare results of NS1 antigen rapid test and ELISA in clinically suspected dengue patients.

Materials and methods: Present study was a comparative study conducted on 100 Patients presented with clinical history of Dengue. At Microbiology Laboratory, serum of all samples was assessed for NS1 detection using antigen Rapid test and ELISA. Sensitivity & specificity values were calculated for NS1 antigen rapid test, compared with ELISA.

Results: Out of 100 serum samples collected from suspected cases of Dengue in and around Anantapuramu, 30 (30%) were positive for ELISA and 28 (28%) were positive for Rapid diagnostic test. Sensitivity & specificity when only NS1 was considered on rapid test kits when compared with ELISA were 93.33%, 98.57%,

Conclusion: It is concluded elisa test was superior in the diagnosis of Dengue and recommended on improvement in sensitivity of RDTs.

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Keywords: NS1 Rapid diagnostic test, NS1 ELISA, Dengue infection, Immunoassays

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INTRODUCTION:

Dengue which is endemic in India is a, a mosquito-borne viral disease caused by infection with dengue virus (DENV 1-4) [1,2] After the beginning of illness which corresponds to the presence of fever, the dengue virus is seen in serum or plasma & tissues of immune system. onset [3] . As there is no particular treatment for dengue, it can be controlled by early detection, providing proper medical care and using proper vector control methods. Currently many laboratories are following the methods like viral isolation, detection of viral genomic sequence by nucleic acid amplification technology assay (RT-PCR), Antigen detection, particularly non-structural protein 1 (NS1) and the detection of dengue virus-specific IgM antibodies by the IgM-capture enzyme linked immunosorbent assay (MAC-ELISA) and/or the rapid dengue immunochromatographic test (ICT) [4].

Immunoassays for NS1 and IgM offer a convenient format for dengue diagnosis, and several enzyme-linked immunosorbent assay (ELISA) and rapid diagnostic tests (RDTs) are commercially available. The present study was conducted to compare results of NS1 antigen rapid test and ELISA in clinically suspected dengue fever patients at a tertiary hospital.

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Material and Methods:

Present study was a comparative study conducted in the department of Microbiology at Government Medical College, Ananthapuram from July 2021 to November 2021. Institutional ethical committee approval was taken prior to start of study and informed consent form was taken from all the participants.

Inclusion criteria:

Patients of all age groups, patients with less than 4 days of fever , clinical symptoms and signs of acute dengue like illness and whom serological diagnosis requested for dengue infection.

Exclusion criteria

Non conclusive reports, patients with more than 4 days of fever, already diagnosed cases of dengue (referred or admitted with dengue positive report)

Patients presented with clinical history of Dengue were selected to do this study. Primary details (age, sex, complaints, medical history) were noted in proforma. A total of 100 patients were advised to go with laboratory testing for confirmation of Dengue. Blood samples of patients who have come to General Medicine & paediatrics OPD with less than 4 days of fever onset were collected and sent to laboratory for NS1 detection. At Microbiology Laboratory, serum of all samples was assessed for NS1 detection using two tests simultaneously. The collected blood sample was centrifuged at 2500rpm for 15 min to obtain serum. The serum has subjected to Rapid Dengue NS1 antigen detection test and NS1 ELISA detection by j mitra kits

Test 1: J.Mitra Dengue NS1 ELISA test for detection of NS1 - Serum sample was assessed according to manufacturer's instructions using ELISA Reader and ELISA Washer.

Test 2: J. Mitra RAPID DENGUE TEST Kit for detection of NS1 - Serum sample was loaded into Rapid Dengue Test kit as per manufacturer's instructions.

The patients who tested positive for NS1 Ag or IgM antibody by ELISA were taken as confirmed cases and to be suffering from acute dengue infection. All the results was analyzed and entered into spread excel sheet. Statistical analysis was done using descriptive statistics.

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Sensitivity & specificity were calculated for NS1Rapid diagnostic test compared with NS1 ELISA test.

Result:

In the present study total 100 samples received in laboratory. Females (51 %) out numbered males (49 %). Most common age groups were 0-10 years (48 %) followed by 11-20 years (18 %) & > 40 years (16%) Table 1

Table 1: Age and gender wise distribution

AGE (years)	MALE	%	FEMALE	%	TOTAL
0-10	24	24	24	24	48
11-20	10	10	08	8	18
21-30	04	4	06	6	10
31-40	06	6	02	2	08
>40	05	5	11	11	16

Out of total 100 samples, samples tested positive by NS1 antigen test were 28 (28%) and samples tested positive by NS1 ELISA were 30 (30 %). Table 2

Table 2: Comparison of result by different diagnostic assays

Diagnostic test (n= 100)	No. of dengue	Males	females	Percentage

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	positive samples			
NS1 antigen Immuno chromatographic test (Rapid Test)	29	17 (17%)	12 (12%)	29%
NS1 ELISA	30	17 (17%)	13 (13%)	30%

Sensitivity, specificity, when only NS1 was considered on RDT kits when compared with ELISA were 93.33%, 98.57%, Table 3

Table 3: Sensitivity & specificity:

	RDT		total
elisa	positive	negative	
positive	28	2	30
negative	01	69	70
total	29	71	100

Discussion:

Dengue has become common in tropical and sub-tropical countries. Epidemics are frequently seen [5] in different parts 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. In addition to difficulties with prevention of dengue, definitive diagnosis of the infection has also proven to be difficult because its symptoms are non-specific, especially in the early, acute stage of the infection. The precise diagnosis of dengue infection can be achieved through viral isolation, viral RNA detection through RT-PCR, but this method is time consuming, costly and not within the reach of even most of the tertiary care hospitals, so its diagnosis is based on the detection of dengue specific antibodies and/or NS1 antigen or ELISA. NS1 (DENV Non-structural protein 1) found in both membrane and soluble forms which is highly conserved. The NS1 antigen is highly specific and detectable in serum from

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days 1 to 9 after fever onset; [6] its sensitivity depends on the type of test used and the time since onset of symptoms (it declines in parallel with viraemia), and is higher in primary than secondary dengue.[7,8] While IgM antibodies level increases rapidly and appears to peak about 2 weeks after the onset of symptoms, then decreases to undetectable levels over 2–3 months. [9]

In our study males were slightly more effected than females. Gupta *et al.;* [10] also reported that males were more commonly affected than females. In our study among 100 suspected Dengue cases, 28% were positive by NSI RDT, 30% were positive by NSI Elisa. In study by Mahesh Kumar et al. [11] among 116 suspected dengue cases, 25% were positive by NS1 ICT, 29.3% were positive by NS1 ELISA.

Conclusion: Our study confirms that elisa test was superior in the diagnosis of Dengue and RDT which needs less expertise and which completes within minutes is useful for early detection of dengue infection,but with a little lower sensitivity which requires an improvement.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

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