Seroprevalence of Dengue in North Karnataka

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Abstract

Introduction - Dengue is endemic in many parts of India and epidemics are frequently reported from various parts of India and abroad. *In India prevalence is more along the East coast, Karnataka, Delhi, Gujarat, northern part of UP. Epidemic outbreaks usually occur during monsoon.*

Aim – *the current study is undertaken to know the Seroprevalence of Dengue infection in North Karnataka.*

Material & Method: *A total of 410 clinically suspected samples were tested for Dengue NS1, IgG, & IgM positivity by rapid test kit.*

Results: *All the 410 samples were subjected to NS1, IgM, IgG Microlisa test. In this positivity to either NS1, IgM, IgG or combined was seen in 45 cases. Our study showed seroprevalence of dengue infection of 10.97%. NS1 Antigen positivity was seen in 20 cases. 21 samples were positive to IgM and 12 samples were positive to IgG.*

Conclusion: *High prevalence rate in our region particularly in post monsoon season gives an alarm to the doctors regarding early and accurate diagnosis of dengue virus infection and its complications. Prompt diagnosis of index cases can facilitate vector control activities in the community so as to mitigate further transmission.*

Keywords: *NS1 Antigen, IgG, IgM*

Introduction

Dengue is a mosquito-borne virus infection prevalent in tropical and subtropical regions around the world, and it has emerged as an important global public health challenge. In recent years, transmission has increased predominantly in urban and semi-urban areas. Dengue is believed to infect 50 to 100 million people worldwide a year with half a million life-threatening infections requiring hospitalization, resulting in approximately 2.5% deaths[1]

Dengue is endemic in many parts of India and epidemics are frequently reported from various parts of India(2–4) and abroad.(5,6) In India prevalence is more along the East coast, Karnataka, Delhi, Gujarat, northern part of UP.

Epidemic outbreaks usually occur during monsoon.

Dengue virus infection produces a spectrum of clinical illness, ranging from an asymptomatic or mild febrile illness to classic dengue fever (DF) to the most severe form of illness, dengue hemorrhagic fever (DHF). [7] There is no specific treatment for dengue/ severe dengue, but early detection and access to proper medical care lowers fatality rates below 1%.[1]

Dengue (DEN) virus belongs to the genus Flaviviruses, and consists of four serotypes: serotype 1 (DEN-1), serotype 2(DEN-2), serotype 3 (DEN-3), and serotype 4 (DEN-4).

Recovery from one type of virus infection, though, provides lifelong immunity against that particular serotype, but there is a strong evidence suggesting the occurrence of severe clinical manifestations of dengue fever in subsequent infection from other serotypes [8]. Infective female Aedesaegypti mosquito species is the primary vector for dengue which transmits the virus through biting humans. On the other hand, Aedesalbopictus is responsible for maintaining the endemicity in the population [9]. Absence of an effective vaccine, vector control measures, and personal protection represent the only available mitigation strategies against dengue outbreaks.

The major diagnostic methods currently available are viral culture, viral RNA detection by reverse transcriptase PCR (RT-PCR) & serological tests such as IgM Capture & IgG Capture ELISA. However early dengue diagnosis still remains a major problem as all these assays have their own pitfalls. The first two assays have restricted scope as 4 a routine diagnostic procedure. Viral isolation by Immunoflourescence though a gold standard cannot be used as a routine diagnostic procedure due to its low sensitivity, laborious procedure & time consumption. The MAC- ELISA which is a commonly used assay has low 5-7 sensitivity in first few days of illness.

Recently, commercially available kits have been developed for the rapid detection of dengue infections. These kits are designed based on the principle of detecting the presence of NS1 antigen and/or anti-dengue antibodies in the blood of suspected dengue patients.

The NS1 is a highly conserved glycoprotein that is present at high concentrations in sera of dengue-infected patients during the early clinical phase of disease, and is found from Day 1 and up to Day 9 after onset of fever in sample of primary or secondary dengue-infected patients. The IgM become detectable on Day 3 to 5 of illness in case of primary dengue infection and persist for 2 to 3 months, whereas IgG appear by the fourteenth day and persist for life. Secondary infection shows that IgG rises within 1 to 2 days after onset of symptoms, simultaneously with IgM antibodies. Therefore, patients with secondary infections will have a positive IgG result, usually, but not always with a positive IgM result.[10, 11, 12] (Fig 1)

Fig 1: Response to IgM, IgG, NS1 Ag during primary and secondary Dengue Infection
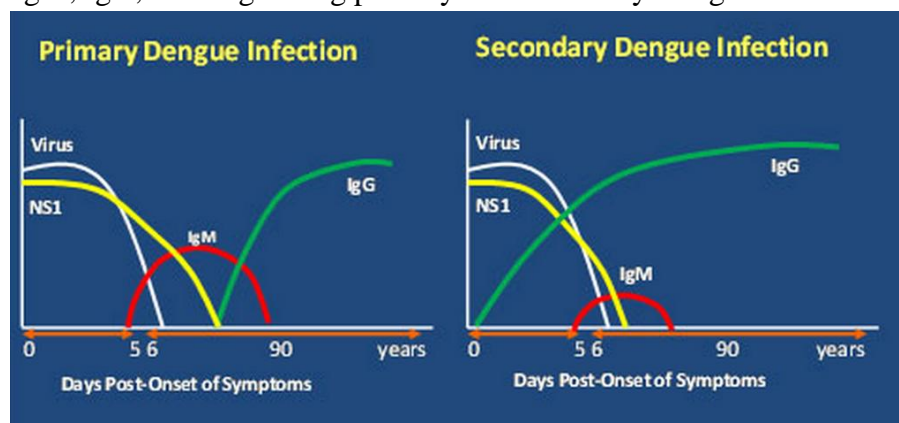
The present communication reports the seroprevalence of dengue infections occurred in North Karnataka by rapid test SD dengue duo kit (IgG, IgM, and NS-1 Ag detection).

Aims & Objectives: The current study is undertaken to know the Seroprevalence of Dengue infection in North Karnataka by using a rapid detection Kit for NS1 Ag, IgM & IgG Antibodies.

Material and Methods

A study was conducted in a tertiary care hospital in north Karnataka considering all clinically suspected samples from May 2014 to April 2015. A total of 410 samples were included in the study. These patients were selected based on a clinical diagnosis of DENV infection and fulfilled the WHO case definition for dengue fever. The status of dengue infection in the patients was determined by serological detection of dengue IgM and IgG, and detection of dengue NS1 antigen.

The detection of Dengue IgG & IgM antibodies & NS1 Antigen was done using Dengue Day 1 kit.

The rapid test kit is produced by and is a one-step immunochromatographic assay designed for the detection of both dengue virus NS1 antigen and differential IgM/IgG antibodies to dengue virus in human whole blood, serum, or plasma. The rapid test contains two test devices; the left side is for dengue NS1 antigen test, whereas the right side is for dengue IgG/IgM test. These kits were designed based on the principle that when a specimen is added to the sample well, anti-dengue IgG and IgM in the specimen will react with recombinant dengue virus envelope proteins-colloidal gold conjugates and forms a complex of antibodies-antigen. This complex will be captured by the relevant anti-human IgG and/or anti-human IgM immobilized on the test device and generate a colored line when migrated along the length of the test device by capillary action. Similarly, dengue NS1 antigen captured by the anti-dengue NS1 Ag-colloid gold conjugate will migrate along the length of the device until being captured by the anti-dengue NS1 antigen immobilized on the membrane strips and generate a color line. All

tests in this study were carried out in accordance with the manufacturer's instructions. Briefly, for dengue NS1 Ag device, 100 µL of the test sample was added into the sample well (S). Test results were interpreted at 15–20 minutes. Similarly, for dengue IgG/IgM device, 10 µL of test sample was added into the sample well (S). This was followed by the addition of 4 drops (90–120 µL) of assay diluent to the round shaped assay diluent well. Results were interpreted at 15–20 minutes. The test results were examined and interpreted according to the manufacturer instructions by three different readers to avoid biasness.

Results

Total of 410 samples from May 2014 to April 2015 were studied. We observed maximum number of clinically suspected cases of dengue in September &October months (Fig -2).

The age group of patients varied from 1-72yrs, with maximum cases between 20-30 yrs. (Fig 3)

All the 410 samples were subjected to NS1, IgM, IgGMicrolisatest. In this positivity to either NS1, IgM, IgG or combined was seen in 45 cases. NS1 Antigen positivity was seen in 20 cases(Early stage of infection). 21 samples were positive to IgM (Primary Dengue infection) and 12 samples were positive to IgG (Secondary Dengue infection). IgG & IgM combined seropositivity was seen in 7 cases indicating secondary Dengue infection. (Table1)

Among the 20 NS1-positive patients there were 7 who were negative for IgM. (table2)

Table 1 : Month wise prevalence of Dengue infection with respect to NS1, IgM, IgG.

Month	Suspected cases	Positive cases	NS1	IgM	IgG	IgM+IgG
May'14	18	1	0	0	1	0
Jun'14	19	1	0	1	0	0
Jul'14	20	1	1	0	0	0
Aug'14	54	5	3	1	2	1
Sept'14	60	12	4	5	4	4
Oct'14	85	14	6	9	2	0
Nov'14	42	5	2	3	2	1
Dec'14	35	3	3	0	0	0
Jan'15	22	1	1	0	0	0
Feb'15	16	0	0	0	0	0
Mar'15	19	1	0	1	0	0
Apr'15	20	1	0	1	1	1

Table 2 : Comparision of NS1 & IgM positivity : To detect early infection

	NS1 +ve	NS1 –ve
IgM +ve	13	8
IgM -ve	7	17

Fig 2 : Month wise prevalence of Dengue : High prevalence rate occurred during September & October Month.
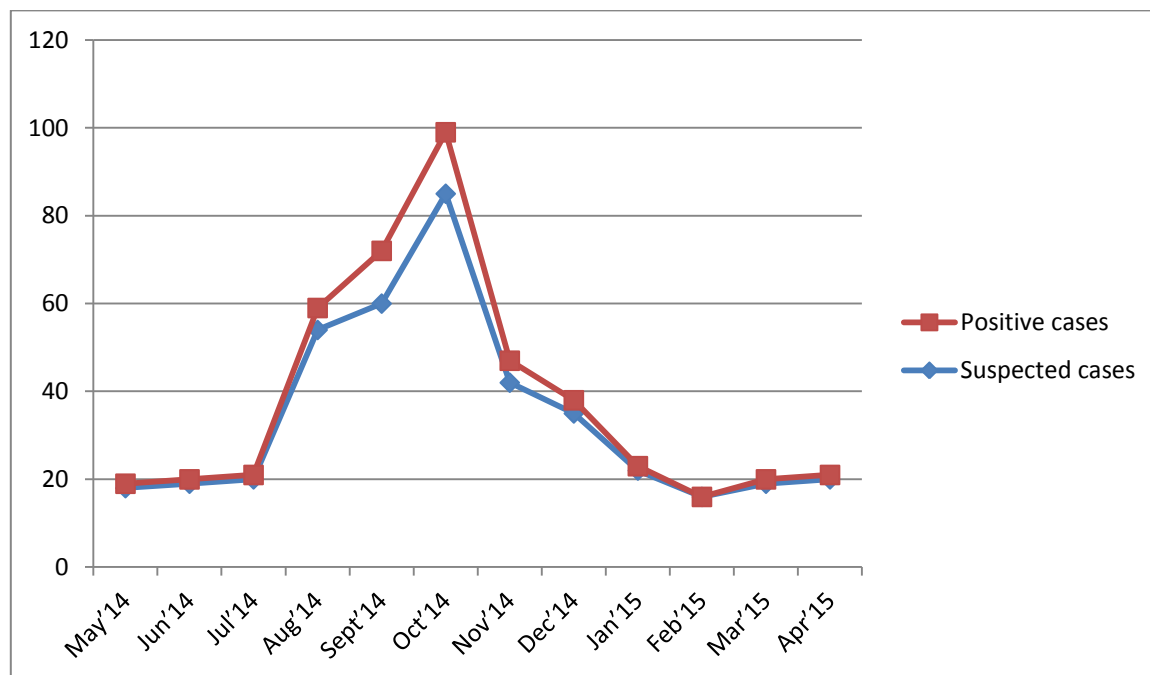
Fig 3 : Age wise distribution of Dengue in suspected & seropositive cases.
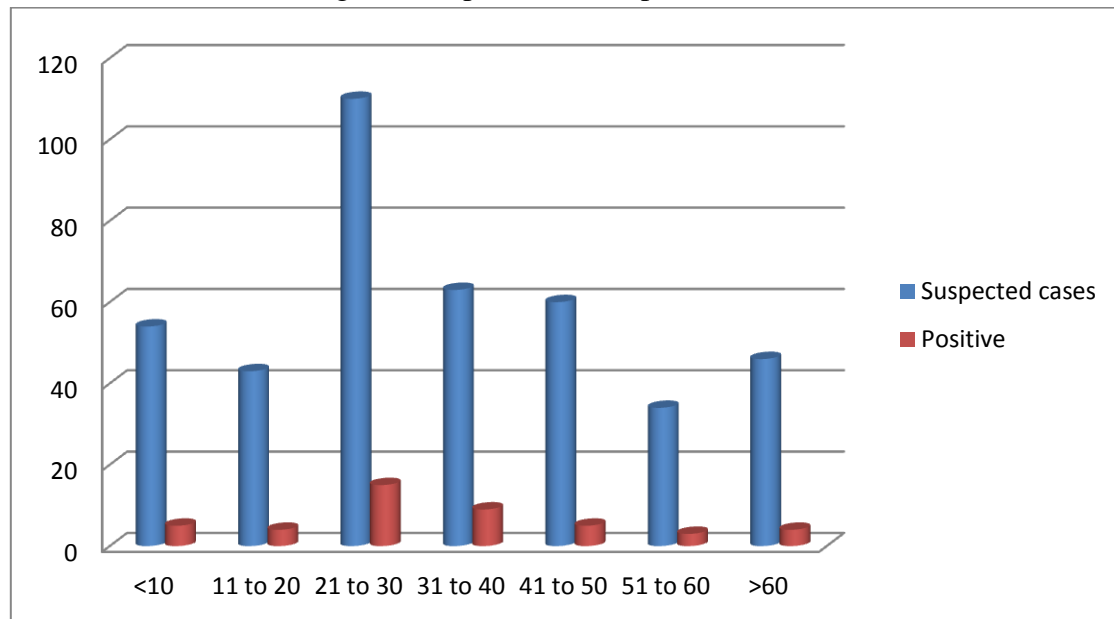
Discussion

Dengue virus infection is the most underrecognized and underreported disease especially in developing countries. This is due to low level of awareness not only among general public but also lack of training to manage dengue among treating physicians, which might result in higher mortality associated with it. In addition, febrile illness is usually confused with other viral diseases and malaria. The commercially available dengue rapid test is suitable for the detection of anti-dengue IgM and IgG antibodies and NS-1 antigen with results available in just 20 mins. Our study showed seroprevalence of dengue infection of 10.97%.

Most of the positive cases 26 out of 45 (57%) occurred during the months of September and october. This is in comparison with similar pattern of month-wise case distribution seen with authors Rasul CH et al in 2002, Narayan et al in 2002, Gomber et al in 2001 , Jayasimha VL et al. 2010 [13, 14, 15, 16] The reasons may be due to the geographical region with prime occupation of the people being agriculture. August & September months are paddy sowing months which needs large stores of water. Also the breeding of Aedesaegypti is highest during pre and post monsoon period. But sporadic cases extend up to

December which indicates endemicity of the infection up to December.

In many parts of the world, dengue infection is predominantly a childhood disease; however, it affects adult population primarily during first few years of its emergence. In India where most affected population stratum in age range of 15–30 years (29.27) [17]. Similar results were seen in our study with maximum cases in age group between 20-30yrs.

Among the 20 NS1-positive patients there were 7 who were negative for IgM [Table 2] and these would have otherwise been missed. They were suffering from a primary infection in the early phase of illness and were also viremic, i.e. they could transmit the virus if bitten by a mosquito. The 12 patients who were exclusively IgG positives, with a secondary viral infection, would have also been overlooked. Without NS1 screening they would have been labelled as "dengue negative". They could have been infectious for mosquitoes during the earlier phase of illness. The concurrent NS1-positive and IgM-positive status of 13 patients [Table 2] reinforced the utility of antigen detection during the earlier phase of illness. The NS1 negatives included 8 IgM positives who had a primary infection presenting a later phase of illness. They were most likely IgG negative. It is unlikely that dengue IgG

antibody levels became undetectable in the convalescent phase of illness.

It would not be possible to rule out Dengue infection in above 365 triple negative cases without testing for viral replication in cell culture or molecular investigation, for which facilities are not available in our hospital. Such patients should be investigated for other acute febrile illnesses.

Conclusion

High prevalence rate in our region particularly in post monsoon season gives an alarm to thedoctors regarding early and accurate diagnosis of dengue virus infection and its complications. Prompt diagnosis of index cases can facilitate vector control activities in the community so as to mitigate further transmission. The commercially available Dengue Day 1 rapid test described in the study should be a valuable screening test for dengue fever. It is rapid, easily be performed,interpreted early and has a extended shelf life. Thestrength of the SD dengue duo rapid test is that dengue IgM and IgG test windows provides additional diagnostic investigation that compliments NS-1 antigen detection.

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