

Detection of Dengue Infection by Rapid IgG/IgM SD Bioline Card Test



Medical Science

KEYWORDS : Dengue, ELISA, IgG, IgM

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ABSTRACT

Background and Objectives: Dengue virus (DENV) is the causative agent of a wide spectrum of clinical manifestations ranging from mild asymptomatic illness to severe fatal dengue haemorrhagic fever/dengue shock syndrome (DHF/DSS). The present study was carried out for rapid diagnosis of dengue infection in children using Dengue Rapid IgM/IgG detection kit. **Materials and Methods:** Serum samples from 58 children <15 yrs of age clinically suspected of having dengue infection and admitted in J K Lone Hospital, Jaipur during the period of two months from November 2011 to December 2011, were tested for the presence of Dengue IgM and IgG antibodies using one-step immunochromatographic assay (Dengue Rapid IgG/IgM Test by SD BIOLINE **Results:** In 15/58 (25.80%) samples, both IgM and IgG antibodies were detected. In 3/58 (5.1%) samples only IgM antibodies were detected whereas in 19/58 (32.70%) samples, only IgG antibodies were detected. In 21/58 (36.20%) samples, none of the antibodies were detected. In total 18/58 (31.30%) samples, IgM antibodies were detected while IgG antibodies were detected in total 34/58 (58.60%) samples. **Conclusion:** Rapid immunochromatographic tests for IgG and IgM offer a convenient method to screen samples for dengue infection in field during epidemic threats. However confirmatory tests are also needed to be done for the confirmation of infection as specificity and sensitivity of these tests has yet to be verified.

Introduction-

Dengue has emerged as a major infectious disease in recent times (Gubler, 1998 and Kyle *et al*, 2008). Dengue virus (DENV) is the causative agent of a wide spectrum of clinical manifestations ranging from mild asymptomatic illness to severe fatal dengue haemorrhagic fever/dengue shock syndrome (DHF/DSS). Worldwide approximately, 2.5 billion people live in dengue-risk areas with about 100 million new cases each year (Gupta N *et al*, 2012). Dengue disease presents highly complex patho-physiological, economic and ecologic problems, especially in disease hyper endemic countries like India. Dengue virus has been prevalent in the Indian subcontinent for the last 50 years (Chakravarty *et al*, 2012) and has become hyper-endemic to dengue with circulation of all the four serotypes (Weaver *et al*, 2009). Dengue affected children are known to have serious disease outcome. After an incubation period of 3-15 days (usually 5 to 8), classical DF begins with an abrupt onset of high fever. During the febrile phase, dehydration may cause neurological disturbances and febrile seizures in young children (WHO guidelines 2009). Recent developments in rapid detection technologies offer the promise of improved diagnostics for case management and the early detection of dengue outbreaks and most of these tests use recombinant antigens from all four dengue virus serotypes and the results are available within 15 to 20 minutes. In the present study, we carried out a rapid diagnosis of dengue infection in children attending SMS and attached group of Hospitals using Dengue Rapid IgM/IgG detection kit.

Materials and methods-

The study was conducted in ICMR Grade I Virology Laboratory, Department of Microbiology and Immunology, SMS Medical College, Jaipur. A total of 58 blood samples from clinically suspected cases of dengue in paediatric age group (1-15 years) admitted in J K Lone Hospital, Jaipur during the period of two months from November 2011 to December 2011 were included in the study. Samples which were haemolysed or having insufficient volume or without proper clinical history were excluded from the study.

Briefly, 2 to 3 ml of blood was collected from each patient using aseptic precautions. Samples were brought to the laboratory maintaining the cold chain. Serum was separated aseptically and stored at 4 °C till tested and at -80°C after use for storage. Samples were simultaneously tested for the presence of Dengue IgM and IgG antibodies using one-step immunochromatographic assay (Dengue Rapid IgG/IgM Test by SD BIOLINE) as per manufacturer's protocol. Briefly 10 µL serum was added to the prescribed sample well (S) followed by the addition of 4 drops (90–120 µL) of assay diluent to the round shaped assay diluent well. The test device is coated with anti-human IgG in the IgG line region and anti-human IgM in the IgM region. When the sample mixture passes through these regions coloured lines appear according to the presence of the antibodies of the serum. The appearance of coloured lines at both regions shows the presence of both IgG and IgM antibodies in the sera. Results were noted within 20 minutes after the addition of the buffer. The control line was observed for the validity of the assay.

Results-

Among children (age<16years) included in the study, mean age of children was 8.1±3.6 years, age ranging from 6 months to 14 years. Table 1 show the results of the one-step immunochromatographic assay which was done for rapid detection of Dengue IgM and IgG antibodies in these samples. In 15/58 (25.80%) samples, both IgM and IgG antibodies were detected. In 3/58 (5.1%) samples only IgM antibodies were detected whereas in 19/58 (32.70%) samples, only IgG antibodies were detected. In 21/58 (36.20%) samples, none of the antibodies were detected. In total 18/58 (31.30%) samples, IgM antibodies were detected while IgG antibodies were detected in total 34/58 (58.60%) samples.

IgG Antibody	IgM Antibody	
	Positive	Negative
Positive	15	19
Negative	3	21

Table 1

38/58 (65.51%) and 20/58 (34.48%) samples were received in the month of November and December respectively, out of which 13/38 (34.21%) and 5/20 (25%) were positive for IgM antibody and 22/38 (57.80%) and 12/20 (75.0%) for IgG (Table 2).

	Total	IgM Pos	IgG pos
Nov-11	38 (65.51%)	13 (34.21%)	22(57.80%)
Dec-11	20 (34.48%)	5 (25%)	12 (75%)
Total	58	18	34

Table 2

Out of 58 cases included, 37/58 (63.79%) were males and 21/58 (32.0%) were females. In IgM positive cases 77.7% were males and 28.5% were females whereas in IgG positive cases 58.8% were males and 41.1% were females (Table 3).

	Male	female
Total samples (58)	37 (63.7%)	21 (36.2%)
IgM positive (18)	14 (77.7%)	4 (28.5%)
IgG Positive (34)	20 (58.8%)	14 (41.1%)

Table 3

All the patients included in the study were suffering from pyrexia and retro-orbital pain (table 4). Other common symptoms were vomiting (67.85%), abdominal pain (50%), arthralgia (15.51%), generalized oedema (10.34%) and seizures (3.4%). 18/58 (31.3%) of total patients had haemorrhagic manifestations and melena was the most common one (17.24%) followed by rash (10.34%), epistaxis (6.89%) and hematemesis (5.1%). Vomiting and abdominal pain were the two most common symptoms in all three categories (positive for both antibodies, only IgM antibodies and only IgG antibodies). Haemorrhagic manifestations were observed in 4/15 (26.6%) cases positive for both antibodies, 1/3 (33.3%) cases of only IgM antibodies positive and 6/19 (31.57%) cases of only IgG positive cases.

Symptoms	Total samples (58)	Both(IgM&IgG) Positive (15)	Only IgM positive (3)	Only IgG positive (19)
Fever with retro orbital pain	58(100%)	15	3	19
Thrombocytopenia	36(62.06%)	12 (80%)	2	13(68.42%)
Vomitting	38(67.85%)	10(66.6%)	2	16
Arthralgia	9(15.51%)	1(6.6%)	1	4
Abdominal pain	29(50%)	11(73.33%)	1	10
edema	6(10.34%)	1(6.6%)	0	1
Resp.distress	1(1%)	0	0	0
seizures	2(3.4%)	1(6%)	0	0
hemorrhagic manifestations	18(31.3%)	4(26.6%)	1	6(31.57%)
Melena	10(17.24%)	4 (26.6%)	0	4
Epistaxis	4(6.89%)	1(6.6%)	0	2
Rash	6(10.34%)	1(6.6%)	1	2
Hemetemesis	3(5.1%)	0	0	1

Table 4

Discussion-

Dengue is the most important arboviral disease and cause severe morbidity and mortality. It presents with a wide spectrum of clinical manifestations and it is often difficult

to predict the clinical evolution of the disease. Dengue can be self-limiting and non-severe or may progress into a severe life threatening hemorrhagic condition characterized by multiple organ failure and plasma leakage. Since there is no treatment for dengue and the treatment is to treat the symptoms, an early diagnosis of dengue becomes critical. An early diagnosis would help treating patients symptomatically at an early stage, thereby preventing progression to a life threatening disease. Since India is hyper endemic to dengue, outbreaks of dengue are frequently reported, almost every year from one part of the country or the other, thereby necessitating for a rapid and reliable test to detect dengue infection. In field, where least laboratory setup is available, a simple and rapid diagnostic test is the method of choice which needs lesser technical expertise. In the present study, we carried out a rapid diagnosis of dengue infection in children attending SMS and attached group of Hospitals using a one-step immunochromatographic assay for rapid detection of dengue IgM and IgG antibodies.

It has been reported that the children are most affected from dengue and the dengue affected children are known to have serious disease outcome (Vajpeyi et al 1999, Garg et al, 2011). Therefore, the present study was conducted in individuals less than 15 years of age

In our study, total 18/58 (31.03%) samples were found to be positive for dengue IgM antibodies that generally appear 3 to 5 days after onset of illness and are indicative of recent dengue infection. In dengue IgM positive cases, majority (63.7%) were males. This reflects that dengue infection rate is higher among male population as reported earlier in other studies also (Gupta et al 2005, Ahmad et al 2008).

Dengue is reported to increase gradually from August, reaches at peak during September and October and then gradually decreases (Vajpeyi et al 1999, Gupta et al 2005, Ukey et al 2010). However in our study Dengue positivity was observed even during November and December.

Fever was observed to be the most common symptom followed by vomiting and abdominal pain in dengue IgM positive cases as found in previous studies (Chairulfatah et al 1995, Ahmed et al 2008). Hemorrhagic manifestations were seen in 5/18 (27.77%) cases of IgM positive cases with gastrointestinal bleeding being the most common (22.22%). Seizures were seen in one case.

In 19/58 (32.70 %) cases, only IgG antibodies were observed. IgG antibodies are reported to appear after IgM antibodies but in case of secondary infection the levels of IgG antibodies show a rapid rise and become detectable at 4-5 days of illness (Shu et al 2003, Sa-Ngasand et al 2006). In present study, 6/19(31.57%) of IgG positive cases had hemorrhagic manifestations. This indicates a probable secondary dengue infection. Since no paired sera were taken from the patients, it was not possible to confirm the infection and this was limitation of this study

Conclusion-

Rapid immunochromatographic tests for IgG and IgM offer a convenient and quick method to screen samples for dengue infection in field during epidemic threats. However confirmatory tests are also needed to be done for the confirmation of infection.

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