



## CORRELATION BETWEEN NS1 WITH PLATELET COUNTS IN DENGUE: AN INSTITUTION EXPERIENCE

**Arti Sharma\***

Resident, MD Pathology, Gajra Raja Medical College, Gwalior, Madhya Pradesh, India \*Corresponding Author

**K S Mangal**

Associate Professor, Department Of Pathology Gajra Raja Medical College, Gwalior, Madhya Pradesh, India

**Rajesh Gaur**

Professor, Department Of Pathology Gajra Raja Medical College, Gwalior, Madhya Pradesh, India

### ABSTRACT

Dengue virus (DENV) infection is one of the mosquito-borne viral diseases with a major impact on public health, globally. Dengue is caused by one of the four serotypes of the dengue virus DENV (DENV-1 to DENV-4) belonging to the family Flaviviridae. Symptoms of manifestation ranged from non-specific viral disease to severe cases characterized by thrombocytopenia, hemorrhage manifestations and hemo-concentration due to plasma leakage. The present were planned with to find correlation between NS1 and platelet count to diagnose early haemorrhagic manifestation.

**KEYWORDS :** Dengue, NS1, Platelet count

### Introduction

Dengue virus (DENV) infection is one of the mosquito-borne viral diseases with a major impact on public health, globally<sup>1</sup>. About 3.5 billion people, 55% of the world's population living in tropical and subtropical regions is at risk, with about 50 million DENV infections occurring annually and approximately 500,000 requiring hospitalization annually<sup>2</sup>.

Dengue is a mosquito-borne viral illness caused by one of the four serotypes of the dengue virus DENV (DENV-1 to DENV-4) belonging to the family Flaviviridae. They are transmitted mainly by the *Aedes aegypti* mosquito and also by *Aedes albopictus*<sup>3</sup>. Dengue has been an urban disease but now has spread to rural areas of India as well<sup>4</sup>.

Dengue virus infection is a complex disease with symptoms being difficult to distinguish from other common febrile illnesses during acute phase and can progress from a mild, non-specific viral disease to severe cases characterized by thrombocytopenia, hemorrhage manifestations and hemo-concentration due to plasma leakage. The present were planned with to find correlation between NS1 and platelet count to diagnose early haemorrhagic manifestation.

### MATERIAL AND METHODS

**Study design:** This study was a hospital-based prospective study conducted for a period of 1 year (May 2015 to April 2016) in Department of Pathology, Gajra Raja Medical college (GRMC) and associated Jayarogya group of hospital (JAH) Gwalior in association with Defense Research and Development Establishment (DRDE) Gwalior. The institutional ethical committee approved the study protocol.

### Participants:

The study group consists of 100 patients admitted in our hospital with dengue Immunoglobulin M (IgM) positive. Patient suffering from any infection, viral hepatitis during this period were excluded from study.

### Case Definition<sup>6</sup>:

Dengue fever: Acute onset of high fever 3–14 days after the bite of an infected mosquito. Symptoms include frontal headache, retro-orbital pain, myalgias, arthralgias, rash, and low white blood cell count.

Dengue haemorrhagic fever: World Health Organization (WHO) criteria:

1. Fever or recent history of fever lasting 2–7 days.
2. Any hemorrhagic manifestation.

3. Thrombocytopenia: Platelet counts less than  $100 \times 10^9/L$
4. Evidence of increased vascular permeability.

Dengue Shock Syndrome: defined as any case that meets the four criteria for DHF and has evidence of circulatory failure manifested by (1) rapid, weak pulse and narrow pulse pressure ( $\leq 20$  mmHg [2.7 kPa]) or (2) hypotension for age, restlessness, and cold, clammy skin.

### Method:

Demographic and clinical characteristics of the participants were recorded on a self-designed semi structured per-forma in all these patients. Just after admission, 5ml. of blood was collected aseptically from each patient. 1 ml of clotted blood was used for Immunoglobulin M (IgM) antibody by mac ELISA; remaining 2 ml of collected blood sample was kept in EDTA vial and was used for haematological investigations by manually as well as by 5 part analyser. Rest 3 ml of clotted blood after separating serum was used for biochemical test and NS1 Antigen. Serum of NS1 Antigen positive patients were further subjected for RT PCR & Real Time PCR using the QIAamp viral RNA mini kit (Qiagen, Germany) and ABI 7500 fast Dx Real time PCR instrument using the dengue specific primer and probes by CDC, USA for virus serotype isolation.

### Statistical analyses

Data were analyzed using SPSS version 20, with a 2-tailed  $\alpha$  level of 5%. Statistical analyses were performed by Chi-square tests analyses. The criterion for statistical significance was set at  $p_{.05}$ , and for statistical trend at  $p_{.10}$ . Given the small sample size, our hypothesis testing must be considered preliminary rather than definitive.

### Results

#### Epidemiological profile of DENV IgM Positive patients:

Total of 409 cases of clinical fever was admitted in various departments during May 2015 to April 2016 in JA group of Hospitals. 100 cases were positive for IgM, prevalence of dengue seropositive was 24.44%. 80 percent were males ( $n=80$ ) and 20 percent ( $n=20$ ) were female. Male: Female ratio was 4:1. Dengue fever was present in 77% while 23% have dengue haemorrhagic fever. We did not found any case of DSS. The age of all patients ranged from 6 month to 69 years. The mean age was 16 years (SD 10.69) and 24.09 years (SD 12.38) for DHF and DF respectively. 35 samples were found positive for NS1 Antigen.

#### Co-relation between NS1 antigen and platelet count:

our study did not showed any correlation between NS1 antigen and platelet level, although these finding were statistically insignificant.

**Table: Correlation between NS1 with platelet count**

Platelet count	NS 1Positive	NS1 Negative	Pearson correlation
Platelet count < 1,00,000	15	8	R=-.116 P=. 249
Platelet > 1,00,000	20	57	
Total	35	65	

### Discussion

Either one or more of two serological markers (NS1, IgM) were positive in 24.44% cases tested, whereas ratio of DF to DHF ratio was 3.34: 1. Various studies have reported a seropositivity ranging from 15.2% (Kulkarni RD *et al*, 2011)<sup>3</sup> to 39.41% (Santosh Tathe *et al*, 2013)<sup>6</sup> for one or more serological markers of dengue. The present study findings appeared to be lower as compared to 31.3% reported by PM ukey *et al*.<sup>7</sup> and higher than 22% reported by Om prakash *et al* 2015<sup>8</sup>. The present findings may be as due to the spatial diffusion of the virus and vector proliferation within the Gwalior region.

In the present study, 35 % specimens were positive for NS1 antigen. Several studies have reported a lesser values of 16% (Shrivastava A *et al*, 2011)<sup>9</sup>, 23.3% ( Datta S *et al*, 2010)<sup>9</sup>, 30% (Kulkarni RD *et al*, 2011)<sup>5</sup> as well as higher value of even 60 % (Santosh Tathe *et al*, 2013)<sup>6</sup> positivity exclusively for NS1 antigen for diagnosis of dengue. NS1 Ag assay is an effective tool for diagnosis of dengue infection, especially within the first four days of illness (Datta S *et al*, 2010)<sup>9</sup>. A recent meta-analysis for NS1-based test as a diagnostic utility for dengue infection supported the use of single NS1-based test with improved sensitivity of detection when combined with an IgM test (Zhang H *et al*, 2014). We did not find any correlation between positive NS1 antigen and low platelet count although our finding were contradictory from other studies reported definitive positive correlation between low platelet count and positive NS1 antigen .

### REFERENCES

- Guzman MG, Halstead SB, Artsob H, Buchy P, Farrar J, Gubler DJ, Hunsperger E, Kroeger A, Margolis HS, Martinez E, Nathan MB, Pelegrino JL, Simmons C, Yoksan S and Peeling RW. (2010). Dengue: a continuing global threat. *Nat Rev Microbiol*. 8: 7-16.
- WHO. Dengue: guidelines for diagnosis, treatment, prevention and control. WHO. (2009). Geneva, Switzerland.
- Sarkar JK, Chatterjee SN, Chakrabarti SK. Virological and serological studies of cases of hemorrhagic fever in Calcutta. *Indian J Med Ref* 1964b, 52:684.
- Arunachalam N, Murty U S, Kabilan L, Balasubramanian A, Thenmozhi V, Narahari D, Ravi A and Satyanarayana K 2004 Studies on dengue in rural areas of Kurnool District, Andhra Pradesh, India; *J. Am. Mosq. Control Assoc.* 20:87–90.
- Kulkarni RD, Patil SS, Ajantha GS, Upadhyay AK, Kalabhavi AS, Shubhada RM, *et al*. Association of platelet count and serological markers of dengue infection- importance of NS1 antigen. *Indian J Med Microbiol* 2011; 29:359–62.
- Santosh Tathe, Chincholkar VV, Kulkarni DM, Nilekar SL, Ovhal RS, Halgarkar CS. A study of NS1 antigen and platelet count for early diagnosis of dengue infection. *Int.J.Curr.Microbiol.App.Sci* 2013.2(12):40-44.
- Basu 167. A, Jain P, Sarkar P, Gangodkar S, Deshpande D, Ganti K, *et al*. Dengue virus infection of SK Hep1 cells: inhibition of in-vitro angiogenesis and altered cytomorphology by expressed viral envelope glycoprotein. *FEMS Immunol Med Microbiol* 2011; 62: 140-7.
- Luplerdlop N, Misse D, Bray D, Deleuze V, Gonzalez JP and Leard K. (2006). Dengue-virus-infected dendritic cells trigger vascular leakage through metalloproteinase overproduction. *EMBO Rep*. 7:1176–1181.
- Datta S, Wattal C. 2010. Dengue NS1 antigen detection: A useful tool in early diagnosis of dengue virus infection. *Indian J Med Microbiol*; 28:107 10.