Journal of Infectious Diseases and Immunity Vol.3(5), pp.85-89, May 2011
Available online at http://www.academicjournals.org/JIDI
ISSN 2141-2375 ©2011 Academic Journals

Full Length Research Paper

Prevalence of dengue among clinically suspected febrile episodes at a teaching hospital in North India

Atul Garg*, Jaya Garg, Y. K. Rao, G. C. Upadhyay and Suman Sakhuja

Department of Microbiology and Pediatrics, G. S. V. M. Medical College, Kanpur. India.

Accepted 6 April, 2011

Dengue is one of the most serious mosquitos borne viral infection mainly affecting tropical and subtropical countries of the world. In absence of specific treatment and vaccine for dengue fever (DF); vector control is the only method by which spread of dengue can be prevented. As effective control and preventive programmes depend upon improved surveillance data, this study was done to report the seroprevalence of Dengue virus infection at Kanpur, North India. The laboratory records of clinically suspected Dengue patients from January 2006 to December 2010 were analyzed retrospectively for demographic features, seasonal variations, and results of IgM and IgG anti dengue antibodies tested by Dengue IGM capture enzyme linked immune sorbent assay (MAC ELISA). A total of 1227 serum samples were analyzed. Out of which 242 samples (19.7%) were found positive for dengue virus infection. Maximum positive cases were seen in 2010 (46.5%). Seasonal trend showed that infection started appearing in august, peaked in October and slowly tapered by December. The most affected age group was 0 to 15 years of age (Pediatric population), followed by 16 to 30 years group and majority of cases were found to be of secondary dengue virus infection (92%). The present outbreak thus emphasizes the need for continuous sero epidemiological surveillance for the timely formulation and implementation of effective dengue control programme.

Key words: Dengue, dengue haemorrhagic fever, IgM antibody capture enzyme linked immune sorbent assay (MAC ELISA), India, vector.

INTRODUCTION

Dengue virus is a positive-strand RNA virus of the Flaviviridae family with 4 distinct serotypes (DV1-4), and is transmitted to humans by several species of the *Aedes* mosquito. It is a major cause of morbidity throughout the tropical and subtropical regions of the world (Gubler, 1998). 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). Population-based studies suggest that asymptomatic infections are the main outcome of dengue virus exposure. However whenever DHF occurs, it is associated with high morbidity and mortality (Martina et al., 2009).

The global prevalence of dengue has grown dramatically in recent decade. The disease is now endemic in more than 100 countries in Africa, the Americas, the eastern Mediterranean, Southeast Asia, and the Western Pacific, threatening more than 2.5 billion people (Dengue, 2010). 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 12,500 to 25,000 deaths (Dengue, 2010). Although dengue has a global distribution, the world health organization (WHO) South-East Asia region together with Western Pacific region bears nearly 75% of the current global disease burden. The South East Asia region is currently experiencing an upsurge in reported cases of dengue in a number of countries, including India, Sri Lanka, and Thailand (Dengue, 2010). The epidemics from India include those from Calcutta (1963),

*Corresponding author. E-mail: atulgargbhu@rediffmail.com. Tel: 0091955936618.

Vishkapattanam (1964), Vellore (1968), Ajmer (1969), Kanpur (1969), Jalore of Rajasthan (1985), Chandigarh (2002), Mumbai (2004), Ludhiana (2007) and more recently from Delhi (1996, 2003. 2006, 2010) (website www.who.com; Pavri et al., 1964; Cherien et al., 1994; Chaturvedi et al., 1972; Kabilan et al., 2003; Padbidri et al., 1973; Balaya et al., 1967; Dar et al., 1999; Gupta et al., 2005).

Early laboratory diagnosis of dengue virus infection is important and is routinely done by serological test. Anti dengue IgM antibodies appears as early as 3 days after infection and remain in circulation for 1 to 2 month. Anti dengue IgG antibodies appears after 1 week, peaks after 2 to 3 weeks and remain lifelong in circulation. Recently, many laboratories are also performing NS1 dengue antigen detection test, and it is showing promising results (World Health Organization, 1997). As effective control and preventive programmes for dengue infection are based upon improved surveillance data, this study was done to report the seroprevalence of dengue virus infection at Kanpur, a major industrial town of North India.

MATERIALS AND METHODS

In this retrospective study, blood samples from the clinically suspected cases of Dengue were reviewed for a period of 5 years from January 2006 to December 2010. Samples were collected from out-patient department as well as patients admitted in different clinical wards of L. L. R. M. Hospital, Kanpur. It is a tertiary care teaching hospital and provides a full range of medical, surgical and super specialty facilities. Processing of samples was done at Department of Microbiology, G. S. V. M. Medical College, Kanpur, India.

Briefly, 2 to 3 ml of blood was collected from each patient by nursing personnel, male orderlies or physicians using strict aseptic precautions and serum was collected using standard methods. All the samples were collected after obtaining the informed consent from the patients and contacts. Serum collected was tested for IgM and IgG anti dengue antibodies by Dengue IGM capture enzyme linked immune sorbent assay (MAC ELISA) and IgG MAC ELISA (Panbio Pty limited, Queensland, Australia). Briefly the procedure was done as follows: 125 µl of peroxidase-labelled antidengue monoclonal antibody conjugate was added in the microwell containing dengue 1 to 4 antigens (antigen plate), resulting in the formation of antigenantibody complex. Within 10 min of addition of conjugate to the antigen plate, 100 µl of 1:100 diluted serum and control were added to another plate (assay plate) containing antihuman IgM antibodies or IgM antibodies attached to microwell test strips.

The assay plate was incubated at 37°C for one hour and then washed. After that, 100 µl of complexed antigen conjugate solution was transferred from the antigen plate to assay plate which was further incubated for one hour. After incubation, the microwells were washed and 100 µl of tetramethylbenzidine/ hydrogen peroxide (TMB/H_20_2) substrate solution was added to each well. After 10 min of incubation at room temperature, stop solution was added to each well and the colour density of the residue (optical density) was read within 30 min at the wavelength of 450 nm. Patients with positive anti dengue IgM were considered positive cases for dengue viral infection. A primary infection was indicated if IgM/IgG index ration was more than 1.2 and as secondary case if this ratio is less than above value (Shu et al., 2003l). All collected data was later on statistically analyzed and presented.

RESULTS

During the five year study period, 1227 serum samples were analyzed. Out of these, 242 samples (19.7%) were positive for dengue virus infection. From 2006 to 2009, dengue fever was moderately low in prevalence, but its prevalence suddenly jumped in 2010 (46.5%) resulting in an epidemic in this region (Figure 1). Seasonal trend showed that there were no positive cases from January to July every year; the infection started spreading in August, peaked in October and slowly tapered by December. The most affected age group was 0 to 15 years of age (Pediatric population), followed by 16 to 30 years group. Interestingly, dengue cases in pediatric patients rose from 25% in 2006 to 43% in 2010. The youngest age showing positive result was 5 months and oldest case was 78 years old. Male to female ratio was 2:1 and majority of cases were of secondary dengue virus infection (92%) (Table 1 and Figure 2).

DISCUSSION

Dengue is an important emerging disease of the tropical and sub-tropical regions today. It is clear that since last decade, dengue have been occurring regularly with periodic surges in a number of cases (Singh, 2007). In this study, 19.7% patients were serologically positive for dengue infection. Upon analyzing the year-wise distribution of dengue cases, an unsteady increase in the number of dengue patients over the past few years was noted. Of the total 390 cases, 181 (46.5%) were reported in the year 2010 whereas only few cases were reported during 2007 to 2009.

However in 2006, of the 400 samples tested, 34 were positive and these findings are in accordance with other studies from India (Padbidri et al., 1973; Balaya et al., 1967; Dar et al., 1999; Gupta et al., 2005). Kanpur is situated in North India on the bank of Ganges River. Thus there are lots of marshy places which provide excellent mosquito breeding places. Further, this may be partially attributed to the rapid unplanned urbanization with unchecked construction activities and poor sanitation facilities contributing fertile breeding grounds for mosquitoes; it is also true that an increase in the alertness among medical fraternity following the initial epidemic and the availability of diagnostic tools in the hospital have contributed to the increased detection of cases.

To identify the seasonal variation of the disease, analysis of the data on monthly basis were done. A gradual increase in cases was noticed from August with a peak in October, during all the five years of the study. The correlation between occurrence of dengue and monsoon season is clearly evident in this study and is further supported by similar findings from Delhi, Ludhiana, Chandigarh and Karachi (Lal et al., 2007; Ratho et al., 2005; Ahmed et al., 2008; Ukey et al.,

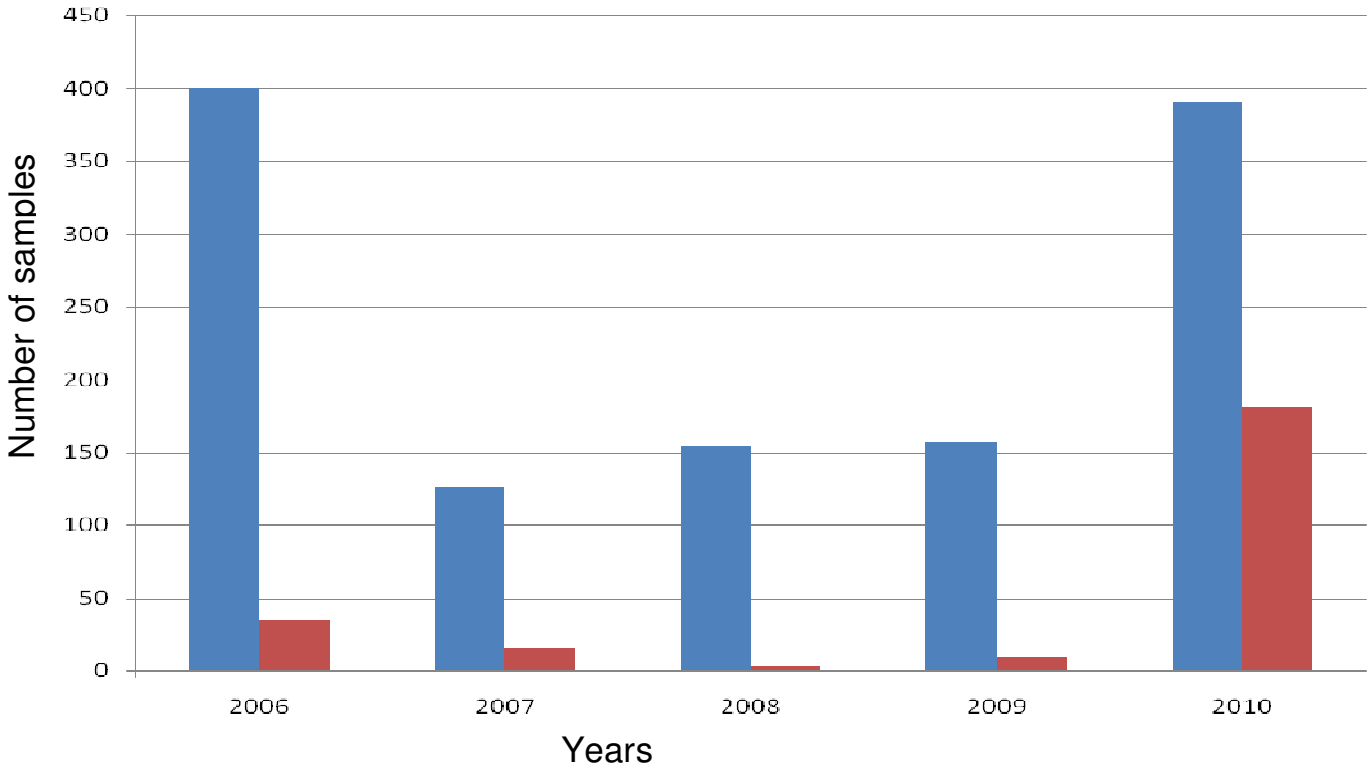

Figure 1. Showing year wise distribution of dengue cases. Blue bar is showing total samples received whereas red bar is showing total positive samples for dengue

Table 1. Showing demographic and immunological characteristics of dengue cases.

Characteristics	Number	Percentage
Age in years		
0-15	103	42.5
16-30	93	38.5
31-45	24	10
46-60	14	5.7
≥ 61	8	3.3
Sex distribution		
Male	162	67
Female	80	33
Immunological type		
Primary infection	18	8
Secondary infection	224	92

2010 Lal et al., 2007; Ratho et al., 2005; Ahmed et al., 2008; Ukey et al., 2010). It may be because this season is very favorable for high breeding of the vector, that is, *Aedes aegypti*. This seasonal outbreak of disease transmission is very important at local level for effective control measures and that preventive measures against dengue infection should come into full swing during water stagnation periods after the initial bouts of rainfall and at the end of monsoon.

The higher prevalence of dengue infection was noted among males than females. The male-to-female ratio was 2:1 which is congruent with other studies (Gupta et al., 2005; Ukey et al., 2010; Kumar et al., 2010). The age group of 0 to 15 years was highly affected with dengue and these finding are not consistent with other Indian studies, as most of the other Indian studies have reported 15 to 45 years as the most affected age group (Gupta et al., 2005; Ukey et al., 2010; Kumar et al., 2010). However in several international studies, dengue has been reported to mainly a pediatric public health problem (Shah et al., 2006; Anderson et al., 2007). It is a very significant finding because true endemicity of dengue is reached when the adult infection declines and only the new entrants into the population, that is, the children, are affected more by the disease. Another very important finding of this study is that among dengue infected patients, only 8% had primary infection whereas majority (92%) of patients had secondary dengue infection. Even secondary dengue infection was seen in infants as young as 6 months and it is a well established fact that complications like dengue haemorrgic fever and dengue shock syndrome occur mainly in cases with secondary infections due to antibody mediated enhancement, Cross reactive T cell response with activation of TH-2 lineage

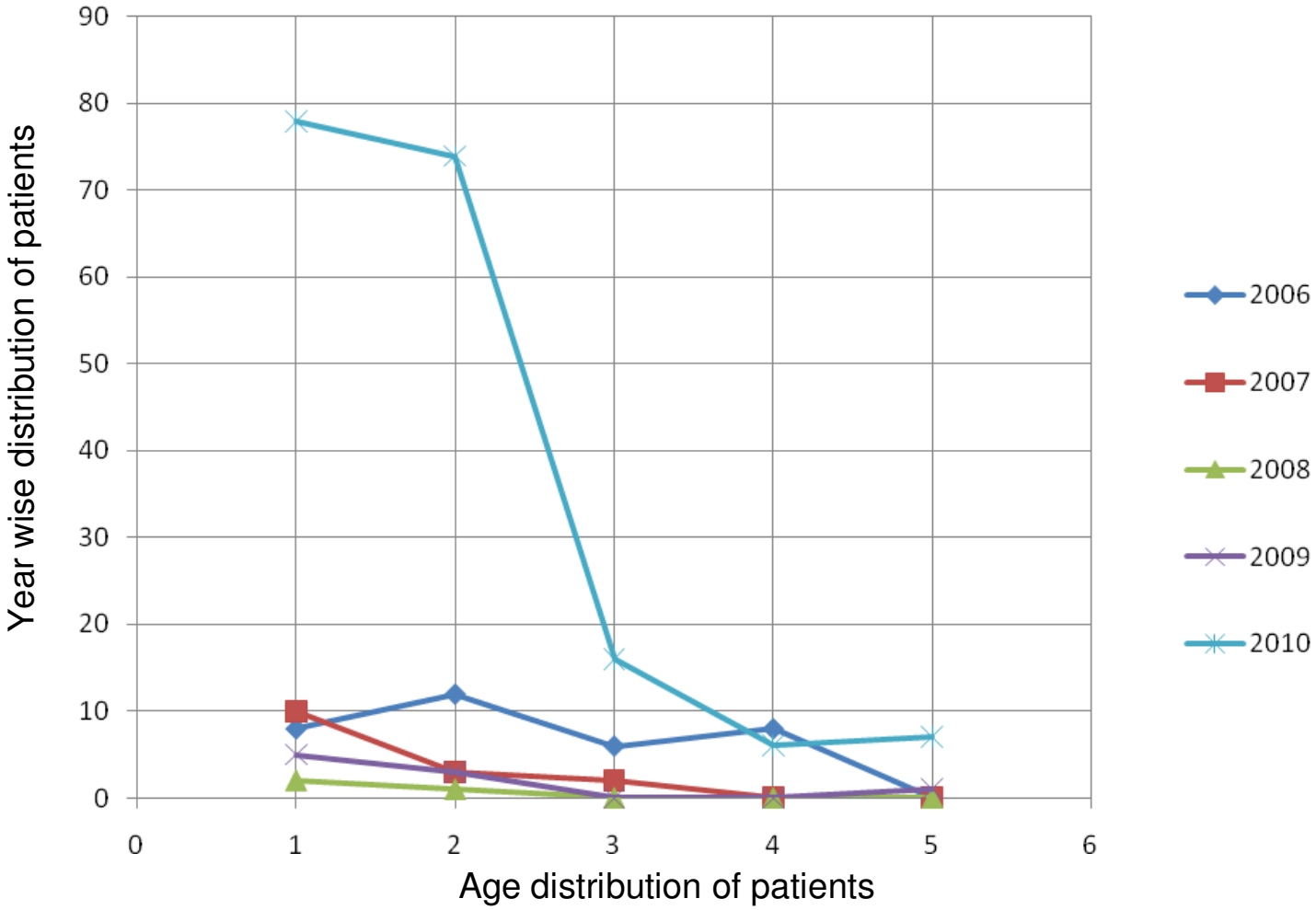

Figure 2. Scatter diagram showing age wise dengue case distribution. Age distribution of patients is as follows: 1 = 0 - 15 years; 2 = 16 - 30 years; 3 = 31 - 45 years; 4 = 46 - 60 years, and 5 = 61 years and above.

cell and stimulation of soluble factors (Martina et al., 2009). The results of this study indicate that dengue infection is not going to wane away but is going to stay and will play havoc if immediate control measures are not taken.

In absence of specific treatment for dengue fever, management is mainly supportive, further there are no vaccines currently available in market thus early diagnosis and vector control is the only method by which dengue can be controlled. Rapid immunochromatograpic test to detect NS1 antigen and IgM antibodies should be available at primary and rural health centers, so that cases can be diagnosed early and thus properly managed. Secondly, the civic agencies have to wake up and rather than adopting a callous attitude and passing the buck, their workers have to 'really work.' Need for enhancing government-citizen partnership through well-coordinated community participation efforts can not be overemphasized. Involving resident welfare associations in urban areas and the Panchayats will help tremendously. The need of the hour is long-term vector control strategy; so that the outbreaks can be prevented and this will simultaneously solve the problem of other mosquito borne diseases like chikungunya, Japanese encephalitis, malaria and filaria.

REFERENCES

Ahmed S, Arif F, Yahya Y, Rehman A, Abbas K, Ashraf S, Akram DS (2008). Dengue fever outbreak in Karachi 2006 - A study of profile and outcome of children under 15 years of age. J. Pakistan Med. Assoc., 58: 4-8.

Anderson KB, Chunsutiwwat S, Nisalak A, Mameen P, Libarty D, Rothman AL (2007). Burden of symptomatic dengue infection in children at primary school in Thailand: a prospective study. Lancet, 369: 1452-59

Balaya S, Paul SD, D'Lima LV, Pavri KM (1967). Investigation on an outbreak of dengue in delhi in 1967. Indian J Med Res ; 57: 767-775

Chaturvedi UC, Mathur A, Kapoor AK, Tandon, Tandon HO, Mehrotra RML (1972). Clinico-virological study of the sequence of dengue epidemic with haemorrhagic manifestations at Kanpur during 1969. Indian J. Med. Res., 60: 329-333

Dar L, Broor S, Sengupta S, Xess I, Seth P (1999). The first major outbreak of dengue hemorrhagic fever in Delhi, India. Emerg. Infect. Dis., 5: 589-90.

Dengue in South East Asian Region (2010). A report by World Health Organization, available on website www.who.com; accessed on 25.2.2010.

Gubler DJ (1998). Dengue and Dengue haemorrhagic fever. Clin. Microbiol. Rev., 11: 480-96

Gupta E, Dar L, Narang P, Srivastava VK, Broor S (2005). Serodiagnosis of dengue during an outbreak at a tertiary care hospital in Delhi. Indian J. Med. Res., 121: 36-8

Kabilan L, Balasubramanian S, Keshava SM, Thenmozhi V, Sekar G, Tewari SC, Arunachalam N,Rajendran R,Satyanarayana K(2003) . Dengue disease spectrum among infants in the 2001 dengue epidemic in Chennai, Tamil Nadu, India. J. Clin. Microbiol., 41: 3919-21

Kumar A, Rao R, Pandit V, Shetty s, Bamigatti C,Samaraging CM (2010). Clinical manifestation and trend of dengue cases admitted in tertiary care hospital, udupi, Karnataka. Ind. Jr. Comm. Med., 35: 386-391.

Lal M, aggarwal A, Oberoi M (2007). Dengue fever, an emerging viral problem in Ludhiana, North India. Ind. Jr. Community Med., 51: 198-199.

Martina BE, Koraka P, Osterhaus A (2009). Dengue virus pathogenesis, an integrated view. Clin. Microbiol. Rev., 22: 564–581.

Padbidri VS, Dandawate CN, Goverdhan MK, Bhat UKM, Rodrigues FM, Lima VD (1973). An investigation of the aetiology of the 1971 outbreak of febrile illness in Jaipur city, India. Indian J. Med. Res., 61: 1737-1743.

Pavri KM, Banerjee G, Anderson CR, Aikat BK (1964). Virological and serological studies of cases of haemorrhagic fever in Calcutta. Indian J. Med. Res., 52: 692-697.

Ratho RK, Mishra B, Kaur J, Kakkar N, Sharma K (2005). An outbreak of dengue fever in Peri Urban slums of Chandigarh, India, with special reference to entomological and climatic factors. Indian J. Med. Sci., 59: 519-27

Shah GS, Islam S, Das BK 2006. Clinical and laboratory profile of dengue infection in children. Kathmandu University Med. J., 13, 40-4.

Shu PY, Chen KK, Chang SF, Yueh YY, Chow L, Chien L J et a (2003). Comparison of capture immunoglobulin M (IgM) and IgG enzyme-linked immunosorbent assay (ELISA) and nonstructural protein NS1 serotype-specific IgG ELISA for differentiation of primary and secondary dengue virus infections. Clin. Diagn. Lab. Immunol., 10: 622–630

Singh B (2007). Dengue outbreak in 2006: Failure of public health system? Indian J. Community Med., 32: 99-100

Ukey PM, Bondade SA, Paunipagar PV, Powar RM, Akulwar SL (2010). Study of seroprevalence of dengue fever in central India. Indian J. Comm. Med., 35: 517-9

World Health Organization (1997): Dengue haemorrhagic fever: Diagnosis, Treatment, Prevention and Control. 2 nd ed, Geneva: World Health Organization, p. 12-23