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A RETROSPECTIVE STUDY TO DETECT DENGUE INFECTION IN UNSUSPECTED FEVER CASES

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ABSTRACT

Dengue is endemic in Kolkata, India. Outbreaks of dengue cases often occur regularly at short intervals. This retrospective seroepidemiological surveillance was conducted longitudinally. Dengue cases were diagnosed in the laboratories from suspected patients by dengue specific IgG, IgM antibodies and NS1 antigen, from suspected persons to investigate, analyze and categorize the cases who were actually suffering from dengue to diagnose a dengue patient in the laboratory practice along with the status of the patient related to the detection of disease and duration of primary and secondary infection for effective monitoring of the patient. Age and Sex of the dengue patients were determined. Detection of dengue in unsuspected fever cases in unfavorable transmission season was evaluated. The transmission of dengue infection in the non-transmission season is not remarkable and often remains submerged. Proper measure at this stage may prevent the epidemic outbreak in the transmission season. This sort of experience will help to enrich the effective control and case management of the menace.

Keywords: Dengue, Non-transmission season, Secondary Dengue cases, Non-suspected dengue cases

INTRODUCTION

Dengue is the most common and vector-borne viral disease. First reported in 1779 by David Bylon during an epidemic in Indonesia, there has been a dramatic expansion in disease distribution in the last 50 years (Hanley & Weaver, 2010). Before 1970, only nine countries had dengue epidemics. Currently, dengue is endemic in more than a hundred countries in five out of the six WHO regions (WHO 2011). It is estimated that there are 2.5 billion people living in dengue-endemic countries (WHO 2015, 2011). Dengue has a wide spectrum of clinical presentations and sometimes its clinical course is unpredictable (WHO 2009). The WHO 1997 guidelines classified dengue into undifferentiated fever, dengue fever and dengue haemorrhagic fever (DHF). DHF was further classified into four severity grades with grade 3 and 4 defined as dengue shock syndrome (DSS) (WHO 1997). However, difficulties in applying the criteria in clinical practice have led to a revision of the classification with the disease classified as severe and non-severe dengue with or without warning signs (WHO 2009). The large cohort is a reflection of the increase in the incidence of dengue in the past decade (Mohd-Zaki *et al.*, 2014).

In the present study, out of 13 cases diagnosed serologically in December, 2011, only one (7.69%) cases were found. During 2012, from January to August, only 25 cases (10.46%) were obtained. Only 1, 0, 0, 1, 1, 1, 8 and 13 cases were recorded from January to August, 2012 respectively (Table 1). These findings indicated that generally in the study area the first six months of the year might be regarded as the non-transmission season of dengue virus. Transmission cycle established in August rapidly expanded in the next three months, as reported in our earlier studies (Hati, 2009).

During this period, when dengue was almost non-existing, many acute febrile cases were referred to the laboratory by the physician, in whom dengue was not suspected, and dengue diagnosis was not sought.

MATERIALS AND METHODS

Study area

Kolkata is one of the large cities of Eastern India and the capital of West Bengal. It has a land area of 185 km2 and a population of about 4.496 million (2011 census). The summer season in Kolkata is from March to June. The rainy season starts in July and ends in October. The

winter season follows from November to February. The annual mean temperature is about 29°C. The annual rainfall is about 1,598 mm. This longitudinal study was conducted in the years 2011-2012.

Data collection

To study the dengue infection in unsuspected fever cases, month-wise distribution dengue cases from December, 2011 to August, 2012 were collected from Gautam Laboratories Imaging and Research Centre, Kolkata, India as computerized data.

Biochemical analysis

The serum samples were tested in the Gautam Laboratories Imaging and Research Centre, Kolkata, India for the presence or absence of dengue specific IgG, IgM antibodies and Ns1 Antigen following the methodology of Guzmán & Kourí, (1996).

To test dengue-specific NS1 antigen, ELISA method was employed using BIORAD kits, MAC ELISA was performed using Vircell kits to detect IgM antibodies. For detection of IgG antibodies BIORAD kits were used. The instructions of the manufacturers were meticulously followed while performing the tests.

RESULTS

A study on 252 non-suspected fever cases

252 serum samples of acute febrile patients, who were not suspected to be suffering in dengue by the referring physicians during December 2011 to August 2012 were analyzed in the laboratory for serological diagnosis of dengue. Each sera sample was tested to detect dengue specific NS1 antigen and IgM and IgG antibodies. Of those 252 persons 145 (57.5%) were male. The age of the patients varied from 4 months to 75 years.

Out of the serum samples collected between December 2011 and August 2012, 1, 1, 0, 0, 1, 1, 1, 8 and 13 dengue cases, totaling 26 (10.3%) were respectively diagnosed (Table 1).

Table 1: Serological diagnosis of dengue infection in fever cases in whom dengue infection was not suspected.

Months	Total Serum samples of patients who are not suspected to be suffering from dengue	Number of confirmed cases of dengue *i.e.* dengue diagnosed
2011, December	13	1
2012, January	29	1
2012, February	23	0
2012, March	22	0
2012, April	33	1
2012, May	23	1
2012, June	14	1
2012, July	30	8
2012, August	65	13
Total	**252**	**26**

Categorization of 26 cases of dengue

Categorization of 26 dengue cases diagnosed from 252 fever patients not suspected to be suffering from dengue infection both dengue-specific antigen NS1 and antibodies IgM and IgG were tested. The results (Table 2) shows (a) early primary (of the three tests only NS1 was reactive and IgG non reactive) one case, (b) early secondary (of the three tests NS1, IgG were reactive) three cases (c) late primary (of the three tests NS1 and IgM were reactive only or IgM reactive and IgG non reactive) five cases and (d) late secondary (NS1, IgM and IgG were all reactive or IgM and IgG only were reactive) 17 cases has been categorized.

Table 2: Categorization of 26 confirmed cases of dengue diagnosed from 252 cases suffering from acute febrile illness and not suspected suffering from dengue

Early primary	Early secondary	Late primary	Late secondary	Total
1	3	5	17	26

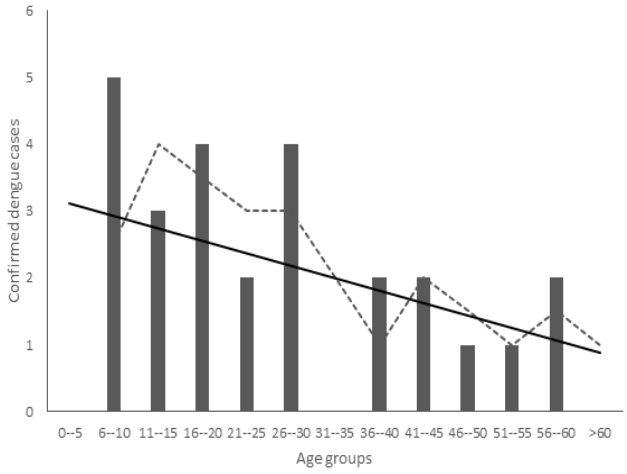
DISCUSSION

Early detection of primary and secondary dengue cases is facilitated by utilizing all three parameters (NS1 antigen, anti-dengue IgM and IgG) which helps in evaluating, monitoring and treating dengue cases effectively (Bhattacharya *et al.*, 2013). Earlier studies showed that antigen capture enzyme-linked immunosorbent assay reveals high levels of the dengue virus protein NS1 in the sera of infected patients (Young *et al.*, 2000).

In current study out of 26 patients 20 cases (76.92%) were secondary dengue cases and it was noteworthy that in this non-transmission season the proportion of secondary dengue cases was found to be very high (Table 2).

The age group of these 26 patients was also studied between 6 and 20 years of age. About 12 (46.2%) dengue cases were found. No dengue was detected above the age of 60 year in this group. A negative association was found between dengue cases and age in the studied population (Figure 1).

Fig 1. Confirmed dengue infection unsuspected fever cases in different age groups reported between December, 2011 and August, 2012. The dotted line shows the moving average model and the solid line shows the linear regression between dengue cases and age group (y = -0.1868x + 3.3077).



Among the 252 non suspected dengue cases no dengue infection was detected in 92 individuals (36.51%) and among these cases 134 individuals (53.17%) suffered from dengue on previous occasion that is they were old dengue cases (134 in 252) (Table 2).

This study pointed out that though the local physician were alert about dengue infection and though they frequently deal with dengue cases in the non-transmission season a sizable proportion of dengue might remain undetected to them.

The smearing nature of dengue infection in so-called non transmission season was probably due to various factors of weather condition limiting the activity of the vector mosquitoes. Therefore it was evident that transmission was going on very insignificantly or in low wave in non-favorable season of the disease. The study of Kassim *et al.*, (2011) showed dengue NS1 antigen test can be used to complement the current antibody test used in peripheral laboratories and the combination of the NS1 antigen and antibody tests could increase the diagnostic efficiency for early diagnosis of dengue infection.

CONCLUSION

The present study advised to give more attention in the non-transmission season to monitor the disease along with control of the vector/vectors and to inhibit rapid spread of the disease in the transmission season. Through the study it revealed that the activity of dengue virus was maintained in the unfavorable season and with the favorable environmental factors outbreak of dengue to the community would occur.

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