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Serological and Epidemiological Picture of Dengue during the Year 2014: An Exclusive Study of Kutch District, Gujarat, India

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The present study was carried out to find out data on Dengue specific serological markers like NS1 antigen, IgM antibody and epidemiological scenario of Dengue cases in Kutch region during the year 2014. Total 765 blood samples from across the Kutch region were tested by ELISA for NS1 antigen and/or IgM antibody as per the protocols and personal, demographic and clinical details of each patient was recorded. Analysis of this data revealed that age group 21-30 years had maximum dengue cases. Male cases (429) were more than females (336). Even though major chunk of Kutch population leaves in rural areas; overall dengue cases were high in urban areas. Out of 765 cases 331 tested positive for dengue. Amongst all positive cases 181(54.68%) were tested positive for NS1 antigen which helped in early diagnosis of dengue. Rise of dengue cases started after the month of August and falls down by the end of December 2014 which coincides with post monsoon season.

Introduction

Dengue is a flavivirus transmitted by Aedes aegypti mosquito. Dengue virus infection has emerged as a notable public health problem in recent decades in term of the mortality and morbidity associated with it (Ukey *et al.,* 2010; WHO, 1997). Dengue is a major problem in many parts of India and large outbreaks are reported from various parts of India and abroad (George *et al.,* 1975; Kaur *et al*., 1997). The clinical picture of dengue mimics many other viral illnesses, which makes laboratory confirmation very crucial for the patient management. The benefits of early diagnosis of dengue fever go beyond

treatment and encase epidemiological efforts. For diagnosis of dengue cases many tests such as virus isolation, RNA, Antigen and antibody detection methods are available (Chakravarti *et al.,* 2006). However virus isolation, RNA detection by PCR, requires well trained staff and dedicated set up which is not feasible in most of the health centers in India. In most of the cases detection of IgM antibody is used for diagnosis of dengue infection. However time required for appearance of IgM antibody is approximately 4-6 days (WHO, 1997). Dengue non-structural 1 antigen (NS1) is highly conserved glycoprotein produced in

both membrane associated and secretary forms is used as a novel biomarker for early diagnosis of dengue infection (Subedi *et al.,* 2014). NS1 antigen detected by ELISA is present in high concentrations in sera of dengue virus infected patients during early clinical phase of disease (Kumarasamy *et al.,* 2007).

Kutch is the largest district of India which has a very special & diverse ecological system. Literature search suggests that there is scarcity of specific & exclusive data on prevalence of Dengue in Kutch region (Madhulika Mistry *et al.,* 2015).

The present study was carried out to find out data on Dengue specific serological markers like NS 1 antigen (Ag), IgM antibody and epidemiological scenario of Dengue cases in Kutch region during the year 2014.

Materials and Methods

This was a prospective & observational study carried out from January 2014 to December 2014 at Department of Microbiology, Gujarat Adani Institute of Medical Sciences, G K General Hospital, Bhuj.

As a designated sentinel surveillance center by National Vector Borne Disease Control Program (NVBDCP), department is receiving serum samples from Primary & Community Health centers as well as affiliated private hospitals from all over Kutch for testing and confirmation of suspected Dengue cases. Patients with acute onset of illness, high grade fever, severe headache, backache, musculoskeletal pain or retro-bulbar pain with or without rashes were considered as clinically suspected case of dengue virus infection (Durani *et al.*, 2014).

There was standard laboratory requisition form with Personal, Demographic and clinical

details which were recorded in same forms. The date of onset of fever and the date of blood sample collection were also recorded in the data entry form. Onset of fever was taken as Day 0 and accordingly sample age was defined as the gap between the date of onset of fever and the date of collection of blood sample.

Serum was separated and then according to the gap between onset of fever and sample collection type of ELISA test was decided. If the gap was 0 to 4 days then NS1 Ag ELISA was performed and if it was more than 6 days then IgM capture ELISA was performed. For the samples having 5 to 6 days of fever both IgM & NS1 Ag ELISA were performed. The Dengue IgM capture ELISA kits were supplied from NIV, Pune under NVBDCP and for NS1 Ag detection Platelia dengue NS1 Ag (Bio-Rad) ELISA kits were used. If any of these test came positive by ELISA it was considered as confirmed case and notified to the concerned government authorities.

Results and Discussion

Total 765 blood samples from across the Kutch region were received at our Institute for serological diagnosis of Dengue fever during the year 2014. Following Figure 1 shows age group wise distribution of clinically suspected and positive dengue cases.

Age group of 21-30 years reported highest number of dengue cases followed by age group of 31-40 years. Figure 2 shows that overall males are more affected then females. Positive cases are also more in males.

From figure 3 it can be observed that 403(52.67%) cases were from urban areas & 362(47.32%) were belonging to rural areas. Positive cases were more from urban areas (46.65%) as compared to rural (39.5%).

Table 1 displays Dengue case presented to the hospital after onset of fever and positive cases amongst them. Majority of cases appeared between 3 to 5 days of fever (325 cases) followed by >5 days of fever (274 cases). Highest positivity was observed amongst the cases that came to hospital within 2 days of fever.

Table 2 shows Number positive cases detected by different ELISA based tests. More than half (54.68%) cases were detected by NS 1 antigen test. While almost 10% of cases were having both the tests, NS 1 Antigen & IgM antibody, positive. Figure 4 depicts month wise trend of Dengue cases for the year 2014. Peak rise of cases starts after the month of August and falls down by the end of December 2014. Another interesting finding is there are small peaks of rise in dengue cases noted between the months of April to July.

Out of 765 suspected cases, highest cases were seen in age group of 21 – 30 years (34.64%) followed by 11 – 20 years age group (22.22%). Also the positive cases amongst both these group showed similar pattern. These show predilection of dengue infection toward young adults and adolescents. These findings are in accordance to other studies by (Durani *et al.,* 2014 & Pandey *et al.,* 2012).

Gender distribution of cases revealed that 429 (56.08%) cases were males and 336(43.92%) were females. Amongst the positive cases 202(61.03%) were males and 129(38.97%) were females. Overall males were more affected which can be due to various factors like their frequent outdoor activities in this region and less reporting of female cases etc. These findings are similar to other studies by Durani *et al*., 2014; Piyush tripathi *et al*., 2008; Nidhi pandey *et al*., 2012.

Table 1 Dengue cases distribution as per days of fever

Number of days between onset of fever and sample collection	Total numbers of sample collected / Suspected Cases (n= 765)	Dengue Positive (n=331)
0 – 2 days	166 (21.7%)	81 (48.8%)
3 -5 days	325 (42.8%)	137 (42.15%)
> 5 days	274 (35.8%)	113 (41.24%)

Table 2 Dengue positive cases by ELISA tests

Test (ELISA Based)	Numbers of Dengue positive Cases (n= 331)	Percentage
NS 1 Antigen	181	54.68 %
NS 1 Antigen + IgM Antibody	32	9.67 %
IgM Antibody	118	35.65 %

Figure 1: Age Group Wise Distribution of Dengue Cases

	0-5	6 - 10	11 - 20	21 - 30	31 - 40	41 - 60	> 60
■ Total Cases	38	43	170	265	133	95	21
■ Dengue Positive	19	13	91	118	42	37	11

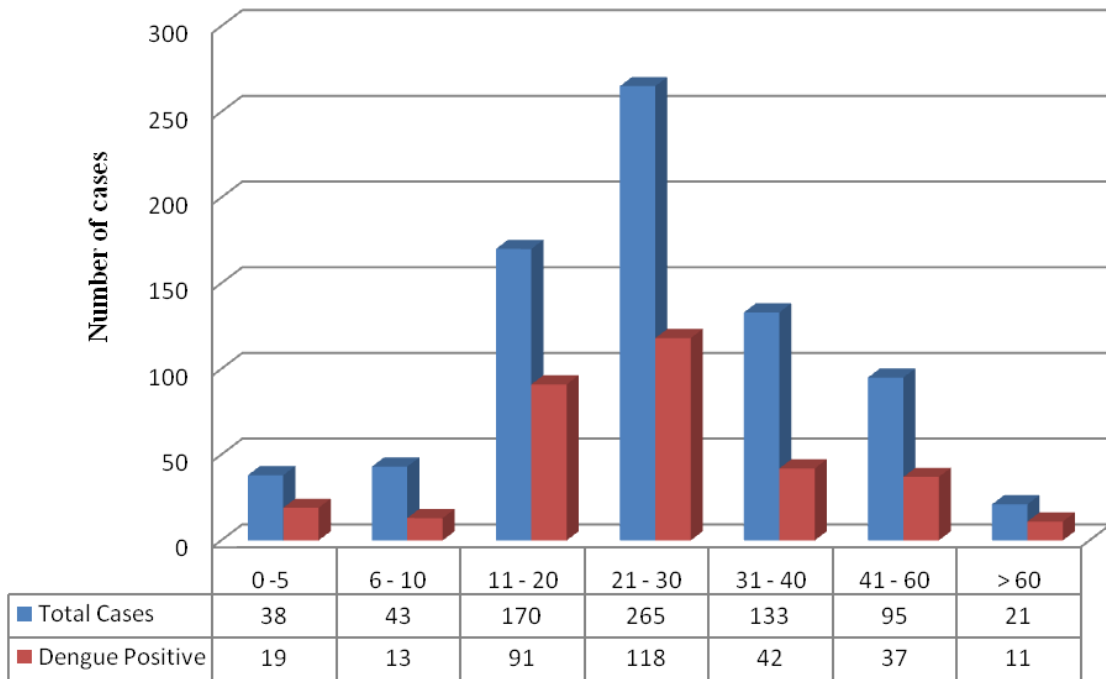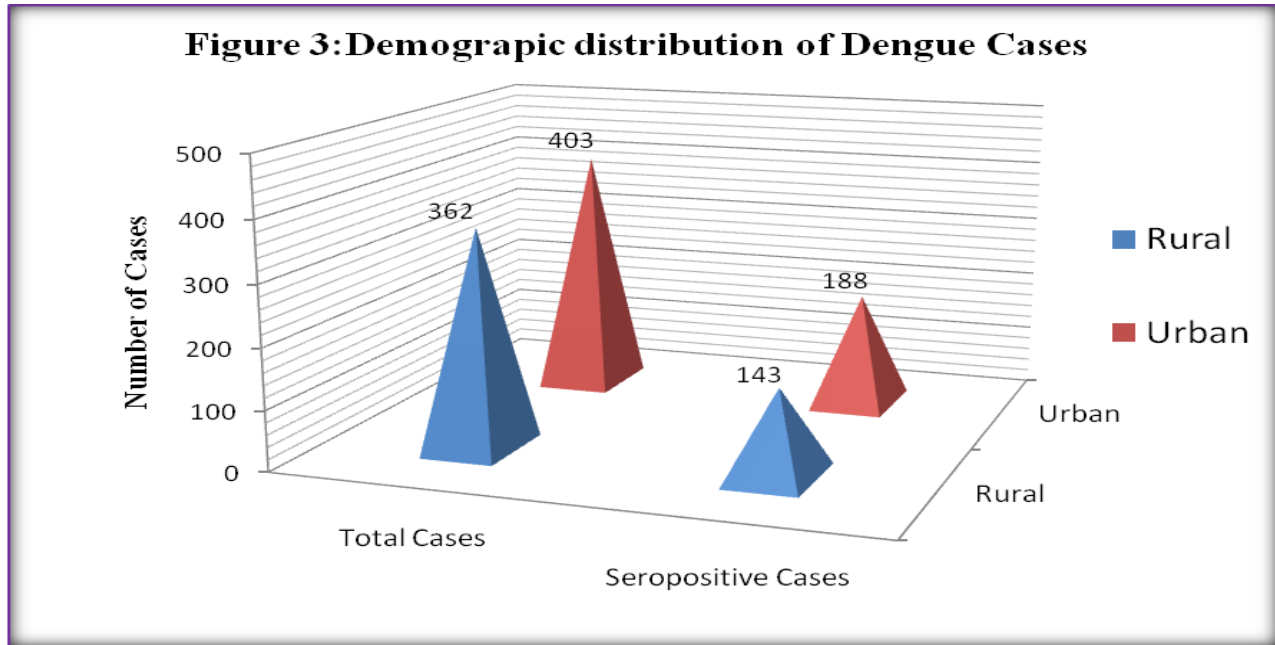## Figure 2: Sex Wise Distribution of Dengue Cases

Figure 3: Demograpic distribution of Dengue Cases

Figure 4: Month Wise Distribution of Dengue Cases Year 2014

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
Total Cases	19	19	10	41	40	29	40	45	139	163	148	72
Positive Cases	7	2	0	15	23	10	20	9	75	73	73	24
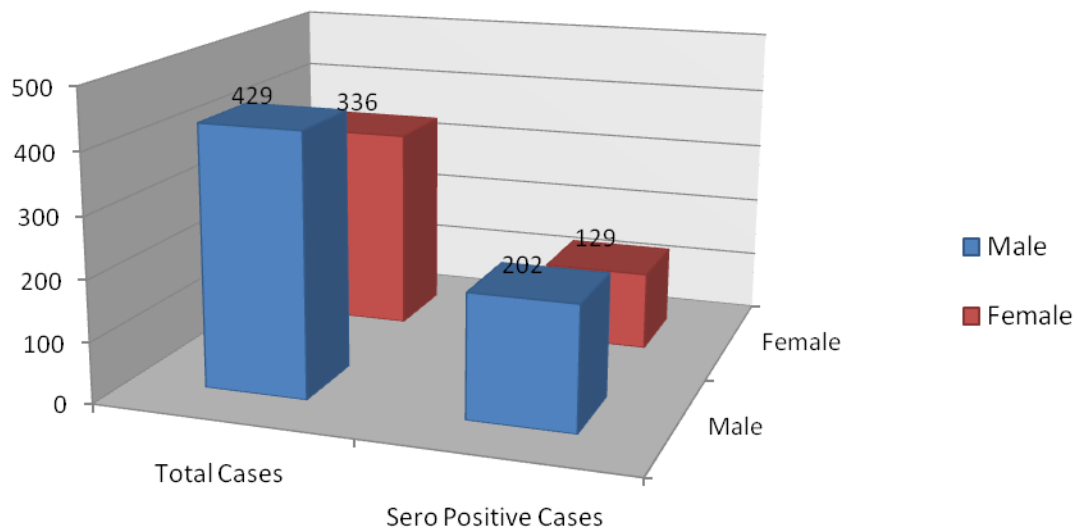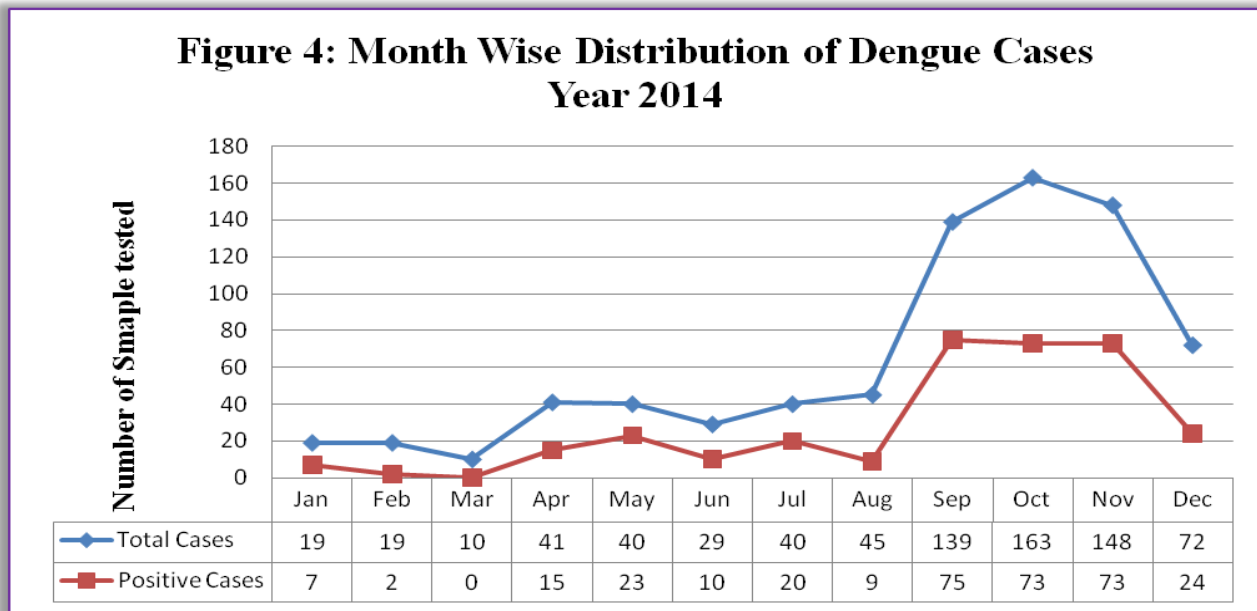
Demographic distribution of Dengue cases in Kutch in 2014 showed that cases from urban areas (403, 52.67%) were slightly higher than the rural (362, 46.65%) areas. Although previous studies from Gujarat & India showed that prevalence of dengue cases were much higher in urban areas; difference here in the present study is much less Madhulika Mistry *et al.,* 2015; Durani *et al*., 2014; Patankar *et al*., 2014; Dutta *et al*., 2012; Khan *et al*., 2014. One reason for this can be natural demography of Kutch region where majority of population lives in rural areas as per Census 2011 data (Government of India, 2011 Census).

In this study testing of samples were done as per the age of sample and it was divided into 3 categories based on gap between onset of fever

and sample collection. As NS1 antigen and IgM antibody levels follow a pattern after the onset of fever; according that the type of test was decided (CDC laboratory guidance for laboratory testing). Maximum number of samples were from 3-5 days of fever (325, 42.48%) followed by >5 days of fever (274, 35.81%). However almost half (81, 48.8%) patients from total 166 patients who came within 2 days of fever were tested positive for Dengue. This could be due to high sensitivity and specificity of Dengue NS 1 antigen ELISA test which was the sole test selected for patients presents early in acute stage of Dengue (Kulkarni *et al.,* 2011; Peeling *et al*., 2010).

Amongst the positive cases (331) detected in this study, more than half (181, 54.68%) were alone detected by NS 1 antigen test followed by Ig M antibody positive cases (118, 35.65%). In rest of 32 positive cases both NS 1 antigen and Ig M antibody were detected. Various studies have indicated that NS 1 antigen helps in early detection and confirmation of diagnosis (Kulkarni *et al.,* 2011; Datta *et al*., 2010; Krunal Mehta *et al*., 2016). When month wise trend was depicted on graph it reinforced the fact that surge in Dengue vector and so the rise in dengue cases occur in Monsoon and post monsoon season (WHO Dengue fever 2nd Edition 1997, Madhulika Mistry *et al.,* 2015; Durani *et al*., 2014; Piyush tripathi *et al*., 2008). Rain starts late in Kutch around end of June and lasts till September. In the year 2014 rise in dengue cases occurred from July; peak reached in October and then started declining. Apart from this; there was unusual small peak of cases during the months of April & May which are hot & dry months. This requires further epidemiological investigation for search of factors which increased dengue transmission in Non seasonal months. Also there is need of long term studies with analysis of dengue serotypes to know more about the epidemiological, demographic and biological variations of dengue cases in Kutch region. It will tremendously help in taking vector control measures and formulating preventive strategies for Dengue in this region.

It is concluded that this study has just opened the first door for analysis of Dengue in Kutch region. It indicates that young adults, both from rural and urban region, are affected more by Dengue. The high number of positive cases during typical post monsoon season indicates failure in vector control steps and alerts clinicians and epidemiologists to be more vigilant for more such upcoming outbreaks. It also emphasizes role of early detection which can be more accurately done by tests like NS 1 antigen and thus helps in implementation of better clinical and preventive measures for entire population at risk.

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