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Original Article

Microbiology Section

Trends in the Seroprevalence of Dengue in a Tertiary Care Hospital of North Karnataka, India

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ABSTRACT

Introduction: Dengue virus of the *Flaviviridae* family is the causative agent of dengue fever. The *Aedes aegypti* mosquito is the main vector for its transmission. Though, the cases of dengue fever are mild and self-resolving, there can be fatal complications like Dengue Haemorrhagic Fever (DHF) and Dengue Shock Syndrome (DSS).

Aim: To study the trends in the seroprevalence of dengue in serum samples of suspected cases.

Materials and Methods: The present study was a cross-sectional study which was conducted from January 2017 to December 2019, at Belagavi Institute of Medical Sciences (BIMS), Belagavi, Karnataka, India. The serum samples were collected from suspected dengue fever cases and tested by Immunoglobulin M (IgM) capture Enzyme Linked Immunosorbent Assay (ELISA), to detect IgM antibody against dengue virus and NS1 capture ELISA for dengue NS1 (nonstructural protein 1) antigen using ELISA kits manufactured by National Institute of Virology (NIV), Pune. The tests were performed according to the manufacturer's instruction. The data obtained from the study was analysed using descriptive statistics.

Results: A total of 8,992 serum samples were tested over a period of three years, of which 1,340 (14.90%) were positive for dengue infection. Among which 1,048 (78.21%) were positive for anti-dengue IgM antibodies, 109 (8.13%) were positive for NS1 antigen and 183 (13.66%) were positive for both. Most affected age group was 11-20 years and male to female ratio was 1.18:1. The seasonal peak was observed in monsoon i.e. month of June (15.52%) followed by August (12.02%).

Conclusion: Seroprevalence of dengue infection being critical signifies the importance of detection of both IgM antibodies and NS1 antigen for diagnosis of dengue infection. The study also identifies younger population being at higher risk and also monsoon as the most favourable season for viral transmission in this region and highlights the importance of concerted efforts towards disease control and prevention.

Keywords: *Flaviviridae*, Immunoglobulin M, Monsoon, Nonstructural protein 1, Paediatric

INTRODUCTION

Dengue, a mosquito-borne infection is caused by a RNA virus of the *Flaviviridae* family. The four distinct serotypes of this virus which cause infections are DENV-1, DENV-2, DENV-3 and DENV-4. The main vector for transmission of dengue is the female *Aedes aegypti* mosquito followed by *Aedes albopictus* [1]. As Dengue is a rapidly spreading tropical disease World Health Organisation (WHO) has declared it a global threat [2]. There has been a drastic increase in the global incidence of dengue in recent years. There are an estimated 390 million infections each year. The number of dengue cases reported to WHO have increased ~6 fold, from <0.5 million in 2010 to over 3.34 million in 2016 [3].

Though most of dengue fever cases are mild and self resolving, it can lead to fatal complications like DHF and DSS [3]. Majority of the cases being asymptomatic during the initial stages of dengue infection, contribute widely to the spread of the disease. Cases of dengue fever usually present with high-grade fever, headache, muscle and joint pains, retro-orbital pain and skin rash. Plasma leakage, haemoconcentration, haemorrhagic shock and multiple organ failure causing mortality is seen in severe Dengue [4]. Probable Dengue Fever (DF) is defined as an acute febrile illness with two or more of the following manifestations: headache, retro-orbital pain, myalgia, arthralgia, rash, haemorrhagic manifestations and leucopenia. Confirmed dengue fever is a case confirmed by laboratory criterion [5].

The diagnostic modalities commonly used for dengue infection are detection of specific antigen and antibodies by ELISA, immunohistochemistry, immunofluorescence, immunochromatographic test, virus isolation, detection of viral RNA by molecular techniques using Nucleic Acid Amplification Technology (NAATs) like Reverse Transcriptase Polymerase Chain Reaction (RT-PCR) [6]. Viral culture and NAATs need specialised and expensive infrastructure and expertise. In systems with limited infrastructure, less expensive tests to detect specific dengue NS1 Antigen, anti-dengue IgM and IgG antibodies can be used for early detection of infection and hence early intervention.

IgM antibodies appear first and can be detected about a week after infection. They peak by 2-4 weeks after the onset of illness. They are found in detectable levels for around three months. IgM detection indicates, a recent infection with dengue. IgG antibodies take longer to develop as compared to IgM. But they remain in the body for years and indicate a past infection [7]. Acute dengue infection can also be diagnosed by detecting the specific NS1 Ag [8]. Dengue NS1 antigen is a highly conserved glycoprotein, produced in both membrane-associated and secretion forms and is detectable in day 1 of fever [9].

Seroprevalence of dengue from North Karnataka region ranged from 6.8% [10] to 45.7% [11] from 2013 to 2019. Thrombocytopenia and leucopenia cases were shown to be as high as 36.26% [12] to 82% [13] and 55.17% [12] to 26% [13] respectively, however there was no mortality. Keeping in view the change in the trends in seroprevalence of dengue with regards to seasonal variations and the passing years, this cross-sectional study was undertaken to analyse the trends in seroprevalence of dengue in a Medical college of a Northern district of Karnataka between 2017 and 2019.

MATERIALS AND METHODS

This cross-sectional study was conducted over a period of three years from January 2017 to December 2019 in Belagavi Institute

of Medical Sciences, Belagavi, Karnataka, India. Ethical clearance was taken from Institutional Ethical Committee (IEC) (BIMS-IEC/93/2019-2020) and informed consent was taken from the patients.

Sample size calculation: Sample size was calculated by referring to the three year study conducted by Shah PS et al., [14].

Inclusion criteria: Patients clinically suspected of dengue infection and who fulfilled the WHO case definition for dengue fever [15] attending Out Patient Department (OPD) and admitted in the wards of Belagavi Institute of Medical Sciences, Belagavi, were included.

Exclusion criteria: Patients with previously diagnosed dengue infection and other proven aetiology of fever (non dengue cases like malaria, tuberculosis, typhoid, etc.) were excluded from the study.

Study Procedure

Blood samples of 8,992 patients having acute febrile illness clinically suspected to have dengue fever [5,12] attending OPD or admitted in the wards were considered. The samples were collected from both, patients who came in acute and in early convalescent stages. Those patients who came with fever of one to four days were considered as in acute stage, while those with fever of more than five days were in early convalescent stage. Most of the samples were tested immediately, using serum obtained after centrifugation of the blood at 3000 revolution per minute (rpm) for 5 min. If immediate testing was not possible, samples were stored at 2-8°C.

Serum samples were tested for IgM anti-dengue antibody and NS1 antigen using IgM antibody capture ELISA kit and NS1 Capture ELISA kit supplied by NIV, Pune (Arbovirus Diagnostic NIV, Pune, India) respectively. The tests were performed following the manufacturer's instruction.

STATISTICAL ANALYSIS

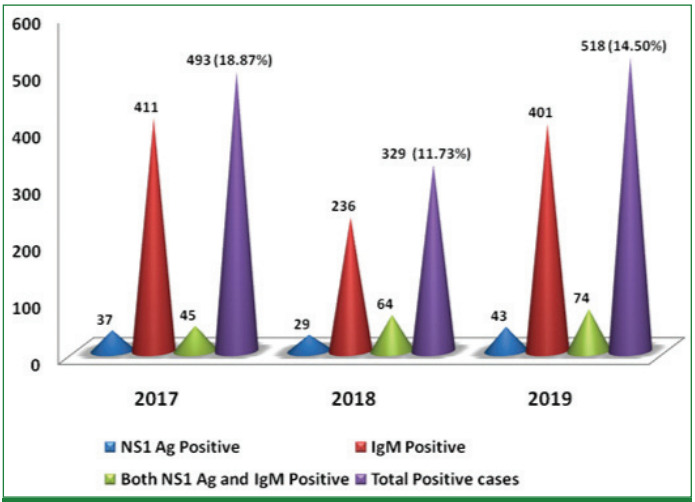
Descriptive statistics was used to analyse the collected data and it was presented in the form of tables and graphs.

RESULTS

A total of 8,992 serum samples (probable dengue cases) were tested over the period of three years, of which 1340 (14.90%) of the samples were confirmed to be positive for dengue infection and 286 (3.18%) were severe dengue cases [Table/Fig-1]. Highest Dengue positive cases were seen in the year of 2017 i.e., 493 of 2613 (18.87%), least being in 2018 i.e., 329 of 2806 (11.73%) [Table/Fig-1]. Among 1340 confirmed cases, 1048 (78.21%) were positive for anti dengue IgM antibodies, 109 (8.13%) were positive for NS1 antigen and 183 (13.66%) were positive for both anti-dengue IgM antibodies and NS1 antigen [Table/Fig-2]. All the age groups were affected, of which the most affected age group was

Dengue cases	2017	2018	2019	Total cases
Probable	2613	2806	3573	8992
Confirmed	493 (18.87%)	329 (11.73%)	518 (14.50%)	1340 (14.90%)
Severe	96 (3.67%)	32 (1.14%)	158 (4.42%)	286 (3.18%)

[Table/Fig-1]: Spectrum of dengue cases over three years.



[Table/Fig-2]: Year-wise seropositivity of dengue cases.

11-20 years with 401 cases (29.93%) followed by 21 to 30 years with 332 cases (24.78%) [Table/Fig-3]. Male to female ratio was 1.18:1. Affected males were 724 (54.03%) and females were 616 (45.97%) [Table/Fig-3].

The maximum number of cases was seen during the monsoon period with highest seasonal peak in the month of June 208 (15.52%) followed by August 161 (12.02%) [Table/Fig-4].

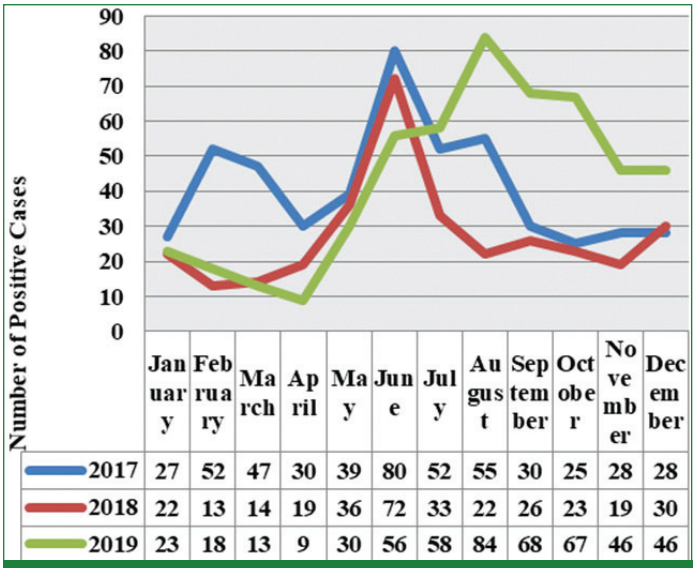
DISCUSSION

Following malaria, dengue fever is ranked the second most prevalent mosquito-borne infection in recent years. Dengue fever cases have reached 40 million and DHF cases are nearing an unsettling figure of several lacks per year [16]. India has been declared as hyper endemic for dengue by WHO and the frequency of cyclical dengue epidemics are found to be increasing here [17]. The reported annual number of dengue cases has been rising steadily [18,19].

This study tested a total of 8,992 serum samples from clinically suspected patients over a period of three years, of which 1340 of the samples were positive for dengue infection accounting for the overall seroprevalence of 14.90%. Similar prevalence was also seen in the studies conducted by Shah PS et al., (15.3%) [14] Vidyasagar K and Venkatesha D (14.33%) [20], Kalita JM et al., (14.85%) [21] and Madkey MV et al., (12.37%) [22]. However, in various other studies conducted

Age group (years)	2017			2018			2019			Total Positive cases (n=1340)		
	Male	Female	Total	Male	Female	Total	Male	Female	Total	Male	Female	Total
0-10	38	64	102	45	31	76	68	34	102	151	129	280 (20.90%)
11-20	80	76	156	40	50	90	60	95	155	180	221	401 (29.93%)
21-30	69	62	131	50	27	77	76	48	124	195	137	332 (24.78%)
31-40	30	24	54	23	15	38	42	19	61	95	58	153 (11.42%)
41-50	12	14	26	15	07	22	30	12	42	57	33	90 (6.72%)
51-60	04	12	16	05	05	10	12	08	24	21	29	50 (3.73%)
61-70	04	01	5	09	05	14	05	01	6	18	07	25 (1.86%)
71-80	02	-	2	-	-	0	02	01	3	04	01	5 (0.37%)
81-90	-	-	0	02	-	2	-	01	1	02	01	3 (0.22%)
91-100	01	-	1	-	-	0	-	-	0	01	-	1 (0.07%)
Total	240	253	493	189	140	329	295	223	518	724 (54.03%)	616 (45.97%)	1340 (100%)

[Table/Fig-3]: Age and gender-wise distribution of dengue seropositive cases over three years.



[Table/Fig-4]: Seasonal distribution of dengue seropositive cases.

Sl. no.	Study	Duration of the study	Place	Seroprevalence of dengue
01	Present study	2017 to 2019	North Karnataka	14.90%
02	Padhi S et al., [6]	2010 to 2012	Odisha	20.05%
03	Umar N and Mir BA [10]	2013 to 2019	North Karnataka	6.8%
04	Kumar MS and Sheethal S, [11]	2014	North Karnataka	45.7%
05	Biradar A et al., [12]	2014 to 2015	North Karnataka	20.42%
06	Shah PS et al., [14]	2014 to 2016	Maharashtra	15.3%
07	Vidyasagar K and Venkatesha D, [20]	2019	South Karnataka	14.33%
08	Kalita JM et al., [21]	2014 to 2018	Rajasthan	14.85%
09	Madkey MV et al., [22]	2018 to 2020	Maharashtra	12.37%
10	Sahu SK et al., [23]	2013 to 2016	Southern Odisha	9.6%
11	Dinakar A and Singh J, [24]	2012 to 2017	Uttar Pradesh	51.22%
12	Islam A et al., [25]	2011 to 2017	South Delhi	58.98%
13	Neralwar A et al., [26]	2014 to 2015	Raipur	32.86%
14	Deasi SS and Desai SV, [27]	2014	Maharashtra	32.72%
15	Mehta KD et al., [28]	2008 to 2011	Gujarat	28%
16	Sathish JV et al., [29]	2017	South Karnataka	20.46
17	Ukey PM et al., [30]	2005 to 2006	Maharashtra	31.3%
18	Goswami L et al., [31]	2013 to 2016	Assam	20%
19	Karoli R et al., [32]	2010	Uttar Pradesh	39%
20	Ahmed NH and Broor S, [33]	2010	Delhi	38.9%

[Table/Fig-5]: Previous trends in seroprevalence of dengue infection [6,10-12,14, 20-33].

in India between 2013 and 2019 the seroprevalence ranges from as less as 6.8% (Umar N and Mir BA) [10], 9.6% (Sahu SK et al.,) [23] to as high as 51.22% (Dinakar and Singh J) [24] and 58.98% (Islam A et al.,) [25]. [Table/Fig-5] shows comparison of seroprevalence of present study with other studies [6,10-12,14,20-33].

Out of 1340 (14.90%) positive cases in the present study, 1048 (78.21%) were positive for anti dengue IgM antibodies, 109 (8.13%) were positive for NS1 antigen and 183 (13.66%) were positive for both anti dengue IgM antibodies and NS1 antigen. Similar pattern was observed in the study conducted by Neralwar A et al., [26] and Desai SS and Desai SV [27]. However in the study conducted by Vidyasagar K and Venkatesha D [20] contrasting results were observed i.e. 73% were positive for NS1Ag and 18% for IgM. The variation in the results observed in different studies can be attributed to the type of serum samples tested whether they are acute or convalescent phase samples and the results of present study signify the importance of testing both IgM antibodies and NS1 Ag to diagnose Dengue infection.

Fluctuating seroprevalence of dengue infection was seen from 2017 to 2019. The decrease in the dengue cases was seen from 2017 to 2018 i.e. 18.87% to 11.73% and again there was a raise in 2019 i.e. 14.50%. Similar trend was also observed in study conducted in Odisha between 2013 and 2016 by Sahu SK et al., [23]. Whereas in most of the studies conducted in India there was a constant rise in the seroprevalence with each year [6,28]. The increase in prevalence in 2019 could be due to the heavy rainfall and flooding during monsoon in 2019 in South India including Karnataka and Maharashtra [34,35].

There was no mortality but morbidity was high as the most affected population belonged to the teenage (11-20 years) and productive age group (21-30 years) followed by paediatric population (0-10 years) 29.93%, 24.78% and 20.90% respectively. The elderly population was least affected. The low prevalence of dengue infection among the extremes of age groups may be because of restricted outdoor exposure and sheltered living. Similar results were observed in studies conducted by Vidyasagar K and Venkatesha D [20] and Padhi S et al., [6]. However in most of the studies, paediatric population was shown to be at higher risk [10,29].

Slight male dominance was seen in the present study with affected male to female ratio of 1.18:1. Similar results were found in studies done by Deasi SS and Desai SV [27], Vidyasagar K and Venkatesha D [20] and Ukey PM et al., [30]. However female dominance was also seen in few studies [6,16,19]. The lesser gender difference in the dengue cases could be due to equal outdoor activities performed by both the genders.

Number of cases was seen to be more during monsoon i.e. month of June (15.52%), August (12.02%) and then July (10.67%). This may be attributed to the increase in vector density during the monsoons and occupational/agricultural activities of the population of Belagavi district. Whereas most of the other studies conducted in India showed that there was a rise in the number of cases after the rainy season with seasonal peak in the month of December (Umar N and Mir BA) [10], October (Mehta KD et al.,) [28] and September (Padhi S et al.,) [6]. However few studies also showed monsoon peak of cases in the month of August [20,29].

The present study was epidemiologically relevant as large number of cases have been tested compared to other studies from this region. Early detection and management was possible as both IgM antibody and NS1 antigen were detected by using ELISA test unlike rapid tests utilised in other studies. Study results were in good correlation with the other studies conducted across the country.

Limitation(s)

Study would have been made more clinically oriented if correlation with other markers like platelet count, leucocyte count and history of transfusions would have been done.

CONCLUSION(S)

The seroprevalence of dengue infection was found to be critical in this study and it also signifies the importance of detection of both IgM antibodies and NS1 Ag for early diagnosis and management of dengue infection. The study also identifies younger population

being most vulnerable for infection and also assessed the seasonal trend of dengue infection and identifies monsoons as the most favourable season for transmission of the virus. There is a need to fortify coordinated vector control measures during the monsoons with focus on personal protective measures among children and young population.

REFERENCES

[1] World Health Organisation (WHO) Dengue/Severe Dengue Available at: www.who.int/mediacentre/factsheets/fs117/en/ (Accessed July 17, 2017)
[2] S. Nebehay. Dengue is fastest-spreading tropical disease, WHO says. 2013. Available from: http://www.reuters.com/article/2013/01/16/healthtropical-idUSL6N0AKCPB2013 0116.
[3] Narayanan M, Aravind A, Ambikapathy P, Prema R, Jeyapaul MP. Dengue Fever clinical and laboratory parameters associated with complications. Dengue bulletin. 2003;27:108-15.
[4] Del Angel RM, Reyes-del Valle J. Dengue vaccines: Strongly sought but not a reality just yet. PLoS Pathog. 2013;9:e1003551.
[5] Sharma Y, Kaur M, Singh S, Pant L, Kudesia M, Jain S. Seroprevalence and trend of dengue cases admitted to a Government hospital, Delhi- 5year Study (2006-2010): A look into the age shift. Int J Prev Med. 2012;3:537-43.
[6] Padhi S, Dash M, Panda P, Parida B, Mohanty I, Sahu S, et al. A three year retrospective study on the increasing trend in seroprevalence of dengue infection from southern Odisha, India, 2014. Indian J Med Res. 140:660-64.
[7] Shah KD, Chithambaram NS, Katwe N. Effectiveness of serological tests for early detection of dengue fever. Sch J App Med Sci. 2015;3(1D):291-96.
[8] Watthanaworawit W, Turner P, Turner CL, Tanganuchitcharnchai A, Jarman RG, Blacksell SD, et al. A prospective evaluation of diagnostic methodologies for the acute diagnosis of dengue virus infection on the Thailand-Myanmar border. Trans Royal Soc Trop Med Hyg. 2011;105(1):32-37.
[9] Young PR, Hilditch PA, Bletchly C, Halloran W. An antigen capture enzyme-linked immunosorbent assay reveals high levels of the dengue virus protein NS1 in the sera of infected patients. J Clin Microbiol. 2000;38:1053.
[10] Umar N, Mir BA. A study on seroprevalence of dengue viral infection using IgM antibody capture ELISA for the early diagnosis in Kalaburagi district, North-Eastern part of Karnataka, India. Int J Med Microbiol Trop Dis. 2019;5(3):138-141.
[11] Kumar MS, Sheethal, S. Comparison of NS-1 antigen detection by ICT and ELISA for evaluating acute dengue. Int J Curr Microbiol App Sci. 2018;7(02):3652-56.
[12] Biradar A, Kauser Y, Itagi I, Jamadar NA. Dengue infection: Its prevalence with seasonal variations. Indian J Microbiol Res. 2016;3(2):89-92.
[13] Ratageri VH, Shepur TA, Wari PK, Chavan SC, Mujahid IB, Yergolkar PN. Clinical profile and outcome of dengue fever cases. Indian J Pediatr. 2005;72(8):705-06.
[14] Shah P S, Alagarasu K, Karad S, Deoshatwar A, Jadhav SM, Raut T, et al. Seroprevalence and incidence of primary dengue infections among children in a rural region of Maharashtra, Western India. BMC Infectious Diseases. 2019;19:296.
[15] Health Organization, Handbook for clinical management of dengue Geneva, Switzerland WHO. 2012.
[16] Raheel U, Faheem M, Riaz MN, Kanwal N, Javed F, Zaidi Nu, et al. Dengue fever in the Indian subcontinent: An overview. J Infect Dev Ctries. 2011;5(4):239-47.
[17] World Health Organization. Comprehensive Guidelines for Prevention and Control of Dengue and Dengue Haemorrhagic Fever. New Delhi;2011.
[18] Mandell, Doughlas and Bennett's principle and practice of infectious diseases, 7th edition, Churchill Livingstone Elsevier, Philadelphia. 2010;2:2133-56.
[19] Shepard DS, Halasa YA, Tyagi BK, Adhish SV, Nandan D, Karthiga KS, et al. Economic and disease burden of dengue illness in India. Am J Trop Med Hyg. 2014;91(6):1235-42.
[20] Vidyasagar K, Venkatesha D. Study of prevalence and serodiagnosis of dengue fever in febrile patients attending a tertiary care hospital. J. Evolution Med. Dent. Sci. 2020;9(21):1637-40.
[21] Kalita JM, Aggarwal A, Yedale K, Gadepalli R, Nag VL. A 5-year study of dengue seropositivity among suspected cases attending a teaching hospital of North-Western region of India. J Med Virol. 2021;93(6):3338-43. Doi: 10.1002/jmv.26592.
[22] Madkey MV, Gedam DS, Meshram VM, Gajbhiye SB. Seroprevalence of dengue in the tribal district of central India. Indian J Microbiol Res. 2021;8(1):45-48.
[23] Sahu S K, Pasupalak S, Mohanty I, Narasimhan M V. Recent trends of seroprevalence of dengue in a tertiary care hospital in Southern Odisha. J Clin Diagnostic Res. 2018;12(1):DC05-DC07.
[24] Dinkar A, Singh J. Dengue infection in North India: An experience of a tertiary care center from 2012 to 2017. Tzu Chi Med J. 2020;32(1):3640.
[25] Islam A, Abdullah M, Tazeen A, Naqvi IH, Kazim SN, Ahmed A, et al. Circulation of dengue virus serotypes in hyperendemic region of New Delhi, India during 2011-2017. J Infect Public Health. 2020;13(12):1912-19. Doi: 10.1016/j.jiph.2020.10.009. Epub 2020 Nov 2. PMID: 33148496.
[26] Neralwar A, Banjare B, Barapatre R. Detection of NS1 antigen, IgM antibody for the diagnosis of dengue infection in patients with acute febrile illness. Int J Res Med Sci 2015;3:2826-30.
[27] Deasi SS, Desai SV. A prospective study on prevalence of intestinal metaplasia and dysplasia in infectious and non-infectious chronic gastritis in a tertiary care teaching hospital. Int J Med Res Prof. 2015;1(3);29-31.
[28] Mehta KD, Gelotar PS, Vachhani SC, Makwana N, Sinha M. Profile of dengue infection in Jamnagar city and district, west India. WHO South-East Asia J Public Health. 2014;3(1):72-74.
[29] Sathish JV, Naik TB, Krishna PVM, Biradar A. Dengue Infection-prevalence and seasonal variation among patients attending a tertiary care hospital at Chamarajanagar, Karnataka, Indian J Microbiol Res. 2018;5(2):275-79.
[30] Ukey PM, Bondade SA, Paunipagar PV, Powar RM, Akulwar SL. Study of seroprevalence of dengue fever in Central India. Indian J Community Med. 2010;35(4):517-19.
[31] Goswami L, Chowdhury R, Rasul ES. Seroprevalence of dengue infection in a tertiary care hospital in Assam. Int J Med and Dent Sci. 2018;7(1):1582-85.
[32] Karoli R, Fatima J, Siddiqi Z, Kazmi K, Sultania A. Clinical profile of dengue infection at a teaching hospital in North India. J Infect Dev Ctries. 2012;6(7):551-54.
[33] Ahmed NH, Broor S. Dengue Fever outbreak in delhi, north India: A clinico-epidemiological study. Indian J Community Med. 2015;40(2):135-38.
[34] "Maharashtra, Kerala, Karnataka Flood Highlights: Nearly 200 Dead In Three States". NDTV. 14 August 2019.
[35] "A single bite: dengue fever hits Karnataka post-monsoon". The New Indian Express, 02nd September 2019.

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