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A study of the clinico-demographical profile of different dengue serotypes circulating in a Southern district of West Bengal, India

Sayani Bose[1], Feradica D Laloo[2], Partha Pratim Maity[3], Anirban Bhaduri[4]

[1]Senior Resident, [3]Assistant Professor, [4]Associate Professor and Head, Department of Microbiology, Tamralipto Government Medical College and Hospital, Purba Midnapore, [2]Senior Resident, Department of Microbiology, Jalpaiguri Government Medical College and Hospital, Jalpaiguri, West Bengal, India

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ABSTRACT

Background: Dengue virus (DENV) which is endemic in many tropical and subtropical areas around the world has four serotypes (DENV1, DENV2, DENV3and DENV4), this viral infection can either cause mild febrile illness Dengue fever or severe, such as Dengue hemorrhagic fever or Dengue Shock Syndrome. The dominant dengue serotypes in a region often change over time and the infecting serotypes are seen to have an impact on the clinical presentation and outcome of the patient. **Aims and Objectives:** Finding the proportion of Dengue non-structural protein 1 (NS1) reactive cases among patients with fever <5 days as well as molecular detection of the prevalent dengue serotypes. **Materials and Methods:** This study was conducted at the Department of Microbiology at Tamralipto Government Medical College and Hospital, East Midnapore, West Bengal. Blood collected from patients suffering from fever <5 days was tested for Dengue NS1. Samples reactive for Dengue NS1 were further tested for dengue serotyping by reverse transcription polymerase chain reaction. The results obtained were analyzed and interpreted. **Results:** Out of 7574 samples of fever patients tested for dengue NS1, 882 samples were reactive and among these 882 NS1 samples, (DENV) ribonucleic acid was detected in 714 samples. The predominant serotype detected was DENV2 (63.73%) followed by DENV3 (26.89%), DENV1 (6.72%), and DENV4 (2.66%). Among the NS1 reactive patients, 52 (5.90%) patients suffered from hemorrhagic complications; among whom the predominant serotype was DENV2 followed by DENV3. **Conclusion:** The infecting dengue serotype seems to have an impact on the disease severity and early detection may help us predict prognosis and need for hospital monitoring, hence reducing complications and mortality. Since the region-wise serotypes are diverse and change every few years, molecular surveillance helps in the early detection of any shift from previously circulating serotypes and thus prepares for handling more severe dengue cases.

Key words: Dengue serotypes; Non-structural protein 1 antigen; Dengue virus

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INTRODUCTION

Dengue, an arthropod-borne viral disease, is caused by Dengue virus (DENV) which belongs to the genus *Flavivirus* of the *Flaviviridae* family and is transmitted to humans through the bite of infected *Aedes* mosquitoes.[1] It is endemic in many tropical and subtropical areas around the world and is considered as one of the major causes of mortality and morbidity in these regions.[2]

DENV has four serologically related but genetically distinct serotypes (DENV1, DENV2, DENV3,

Address for Correspondence:
Dr. Anirban Bhaduri, Associate Professor and Head, Department of Microbiology, Tamralipto Government Medical College and Hospital, Purba Midnapore, West Bengal, India. **Mobile:** +91-9073673120. **E-mail:** anir8365@gmail.com

DENV4) and infection with them can either lead to a mild febrile illness called Dengue fever or a severe form of the disease, such as dengue hemorrhagic fever (DHF)/dengue shock syndrome (DSS).[2,3] A dengue patient, when infected with one serotype, confers lifelong immunity against that same serotype but does not confer strong immunity against infection with other serotypes. DHF/DSS results from immune enhancement after a second infection with a heterologous DENV serotype and can lead to fatality.[4]

The predominant serotypes reported from different parts of India during 1982–2015 were DENV2 and DENV1 in the northern region, DENV2 and DENV3 in the southern region, DENV1 in the eastern region and DENV2 in the western regions.[5] However, the dominant dengue serotypes in a region often keep changing over time.[6] The infecting DENV serotypes are seen to have an impact on the clinical presentation and outcome of the patient.[7,8]

Therefore molecular detection of serotypes in dengue patients could be applied for seroepidemiological study of DENV infection[9] and will also help in identifying any shift from the previously circulating serotypes. This, in turn, could be an early indicator for preparing to handle more severe dengue cases which may be of immense public health importance. Thus, the present study was performed in the district of East Midnapur, West Bengal to identify the different dengue serotypes circulating among the population.

Aims and objectives

Finding the proportion of Dengue NS1 reactive cases among patients with fever <5 days as well as molecular detection of the prevalent dengue serotypes.

MATERIALS AND METHODS

This is a hospital-based observational cross-sectional study performed at the Department of Microbiology at Tamralipto Government Medical College and Hospital, East Midnapore, West Bengal, India for a period of 1 year from April 2023 to March 2024.

Blood collected from patients with fever <5 days was received from all over the district of East Midnapore at our laboratory and tested for Dengue non-structural protein 1 (NS1) using DENGUE NS1 Ag MICROLISA ELISA kit (J.Mitra). To detect the circulating dengue serotypes from the NS1 positive samples, ribonucleic acid (RNA) was extracted using QIA amp Viral RNA Mini kit (QIAGEN), and the extracted RNA was amplified by real-time reverse transcription polymerase chain reaction (RT-PCR) using the ALTONA RealStar Dengue Type RT-PCR Kit (ALTONA Diagnostics).

Results obtained were tabulated, interpreted, and analyzed in Microsoft Office Excel worksheet and SPSS VERSION 9.0 analysis software.

RESULTS

Out of 7574 samples of fever patients tested for dengue, 882 samples were NS1 reactive and among these 882 NS1 samples, DENV RNA was detected in 714 samples. The predominant serotype detected was DENV2 (63.73%) followed by DENV3 (26.89%), DENV1 (6.72%), and DENV4 (2.66%) (Figure 1).

The age group-wise case distribution for each dengue serotype has been shown in Table 1.

The adult age group was the most commonly affected in our study followed by the adolescent and pediatric age groups and then the geriatric age.

Males were more affected than females, the male: Female ratio in our study was found to be 1.6:1. Dengue reactive cases were observed throughout the study period with an upsurge of cases during the months of September, October, and November as shown in Figure 2.

Out of 882 dengue NS1reactive cases, 557 (63.15%) patients received treatment at home, 259 (29.37%) were treated in hospital wards, and 66 (7.49%) patients needed admission in an intensive care unit (ICU) as shown in Table 2. Out of the 52 (5.90%) NS1 reactive patients who suffered from hemorrhagic complications; 38 73.08%) patients were DENV2, 12 (23.07%) were DENV3, 1 (1,92%) was DENV1 while for one patient no serotype was found as shown in Table 3.

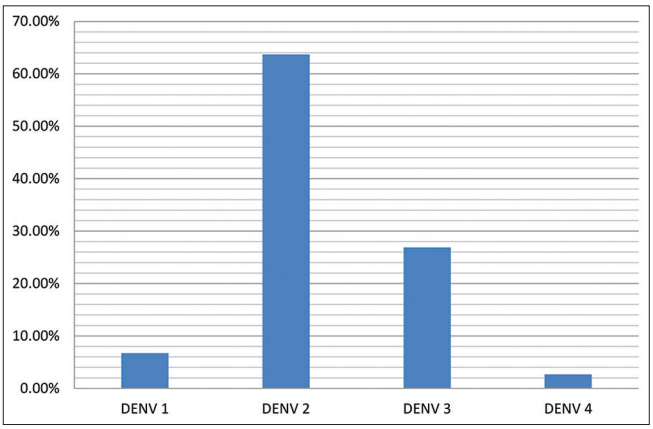

Figure 1: Occurrence rate of different dengue serotypes

Table 1: Age-wise distribution of serotypes

Dengue serotypes	Total number of cases	0–12 years	13–20 years	21–40 years	41–60 years	>60 years
DENV 1	48	3	22	15	5	3
DENV 2	455	30	90	141	153	41
DENV 3	192	25	63	55	34	15
DENV 4	19	2	6	5	5	1

DENV: Dengue virus

Table 2: Treatment facility availed by dengue patients

Treatment	Number of patients	Percentage
OPD	557	63.15
IPD	259	29.37
ICU	66	7.49

OPD: Outpatient department, IPD: In-patient department, ICU: Intensive care unit

Table 3: Dengue serotypes observed in hemorrhagic patients

Serotypes	Number of patients	Percentage
DENV 1	1	1.92
DENV 2	38	73.08
DENV 3	12	23.07
DENV 4	0	0

DENV: Dengue virus

DISCUSSION

Among the 882 NS1 reactive patients, the DENV serotype was detected only in 714 samples. The reason could be because the window for viral RNA detection is limited to 2–3 days after onset of illness and patients often arrive at the hospital after 3 days of onset of fever. All four serotypes were detected throughout the study with a predominance of DENV2 (63.73%) followed by DENV3 (26.89%) serotypes which is a similar finding to several other studies in India.[2,10-13] However few studies have shown different findings such as studies by Bharaj et al.,[14] and Barde et al.,[7] where DENV 3 was found to be the predominant circulating serotype as well as studies by Changal et al.,[15] and Patil et al.,[16] where DENV 1 was the predominant serotype. In a study conducted at ICMR-National Institute of Virology Pune, Alagarasu et al., found that regional diversity was observed with regard to the dominant circulating serotypes in India in the year 2018. This explains the diversity of serotypes found.[17]

The adult age group was the most commonly affected in our study followed by the adolescent and pediatric age group and then the geriatric age which is in concordance with a study by Racherla et al.,[18] The reason for this might be due to Dengue being spread by mosquito bites, therefore people in crowded areas like workplaces and colleges are more likely to be affected.

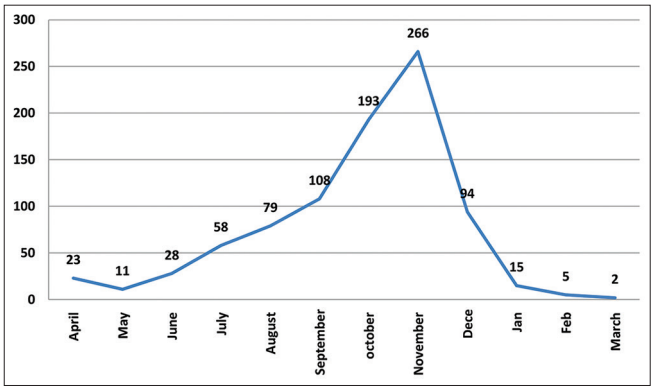

Figure 2: Month-wise distribution of dengue non-structural protein 1 case

In our study a male preponderance was observed among the dengue NS1 reactive cases which might be explained by exposure of males to more outdoor activities, be it occupational or recreational, in a conservative Indian society.[15,19]

Analyzing the month-wise distribution of dengue cases identified, a seasonal upsurge of cases from September to December with a peak in the month of November, similar trends have been observed in other studies done in West Bengal.[2,20] The correlation of increased Dengue cases with monsoon and post-monsoon season can be explained by the fact that stagnant fresh water during the rainy seasons favored the breeding of vector mosquitoes.

While most (63.15%) of the dengue patients recovered at home, few (29.37%) required hospital admission and 7.49% of the patients had to be treated in the ICU. 5.90% of the patients suffered hemorrhagic complications among whom the predominant dengue serotypes observed were DENV2 (73.08%) and DENV3 (23.07%). Various previous studies have suggested that DENV2 and DENV3 serotypes are associated with more severe disease and a higher percentage of DHF cases[21-24] which matches the findings of our study. However, in a study by Yung et al., in Singapore in 2015, it was observed that DHF was more associated by serotype 1 rather than serotype 2.[25] Even though many studies have shown a correlation of the severity of dengue infection with DENV 2 and DENV 3 serotype, further studies are required to establish a definite link between distinct serotypes and severe disease manifestation.[10]

Limitations of the study

This study was done in a resource limited settings and only data of one year was taken in the study. It would be a better study if the circulating serotyping pattern of Dengue virus was taken for a continuous stretch of at least 3 years.

CONCLUSION

The infecting dengue serotype seems to have an impact on the disease severity and early detection may help us predict prognosis and the need for hospital monitoring hence, reducing complications and mortality. The serotype detected region-wise is diverse and changes every few years, therefore surveillance of molecular serotypes and early detection of any shift from the previously circulating serotypes will be an early indicator for the preparedness of handling more severe dengue cases which may be of immense public health importance.

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REFERENCES

1. Wang WH, Urbina AN, Chang MR, Assavalapsakul W, Lu PL, Chen YH, et al. Dengue hemorrhagic fever - A systemic literature review of current perspectives on pathogenesis, prevention and control. J Microbiol Immunol Infect. 2020;53(6):963-978.
   https://doi.org/10.1016/j.jmii.2020.03.007
2. Sarkar A, Taraphdar D and Chatterjee S. Molecular typing of dengue virus circulating in Kolkata, India in 2010. J Trop Med. 2012;2012:960329.
   https://doi.org/10.1155/2012/960329
3. Murugesan A, Aridoss D, Senthilkumar S, Sivathanu L, Sekar R, Shankar EM, et al. Molecular diversity of dengue virus serotypes 1-4 during an outbreak of acute dengue virus infection in Theni, India. Indian J Med Microbiol. 2020;38(3 & 4):401-408.
   https://doi.org/10.4103/ijmm.IJMM_20_89
4. Halstead SB. Pathogenesis of dengue: Challenges to molecular biology. Science. 1988;239(4839):476-481.
   https://doi.org/10.1126/science.3277268
5. Balasubramanian S, Chandy S, Peter R, Nachiyar GS, Sudhakar A, Sumanth A, et al. Utility of a multiplex real-time polymerase chain reaction for combined detection and serotyping of dengue virus in paediatric patients hospitalised with severe dengue: A report from Chennai. Indian J Med Microbiol. 2020;38:288-292.
   https://doi.org/10.4103/ijmm.IJMM_20_249
6. Ganeshkumar P, Murhekar MV, Poornima V, Saravanakumar V, Sukumaran K, Anandaselvasankar A, et al. Dengue infection in India: A systematic review and meta-analysis. PLoS Negl Trop Dis. 2018;12(7):e0006618.
   https://doi.org/10.1371/journal.pntd.0006618
7. Barde PV, Shukla MK, Joshi P, Sahare L and Ukey MJ. Molecular studies on dengue viruses detected in patients from Central India. Indian J Med Microbiol. 2019;37(1):12-18.
   https://doi.org/10.4103/ijmm.IJMM_18_377
8. Vicente CR, Herbinger KH, Fröschl G, Malta Romano C, de Souza Areias Cabidelle A and Cerutti Junior C. Serotype influences on dengue severity: A cross-sectional study on 485 confirmed dengue cases in Vitória, Brazil. BMC Infect Dis. 2016;16:320.
   https://doi.org/10.1186/s12879-016-1668-y
9. Shu PY, Chen LK, Chang SF, Yueh YY, Chow L, Chien LJ, etal. 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. 2003;10(4):622-630.
   https://doi.org/10.1128/cdli.10.4.622-630.2003
10. Kumaria R. Correlation of disease spectrum among four Dengue serotypes: A five years hospital based study from India. Braz J Infect Dis. 2010;14(2):141-146.
11. Gupta A, Rijhwani P, Pahadia MR, Kalia A, Choudhary S, Bansal DP, et al. Prevalence of dengue serotypes and its correlation with the laboratory profile at a tertiary care hospital in Northwestern India. Cureus. 2021;13(5):e15029.
    https://doi.org/10.7759/cureus.15029
12. Shrivastava S, Tiraki D, Diwan A, Lalwani SK, Modak M, Mishra AC, et al. Co-circulation of all the four dengue virus serotypes and detection of a novel clade of DENV-4 (genotype I) virus in Pune, India during 2016 season. PLoS One. 2018;13(2):e0192672.
    https://doi.org/10.1371/journal.pone.0192672
13. Vinodkumar CS, Kalapannavar NK, Basavarajappa KG, Sanjay D, Gowli C, Nadig NG, et al. Episode of coexisting infections with multiple dengue virus serotypes in central Karnataka, India. J Infect Public Health. 2013;6(4):302-306.
    https://doi.org/10.1016/j.jiph.2013.01.004
14. Bharaj P, Chahar HS, Pandey A, Diddi K, Dar L, Guleria R, et al. Concurrent infections by all four dengue virus serotypes during an outbreak of dengue in 2006 in Delhi, India. Virol J. 2008;5:1-5.
    https://doi.org10.1186/1743-422X-5-1
15. Changal KH, Raina AH, Raina A, Raina M, Bashir R, Latief M, et al. Differentiating secondary from primary dengue using IgG to IgM ratio in early dengue: An observational hospital based clinico-serological study from North India. BMC Infect Dis. 2016;16(1):715.
    https://doi.org/10.1186/s12879-016-2053-6
16. Patil JA, Alagarasu K, Kakade MB, More AM, Gadekar KA, Jadhav SM, et al. Emergence of dengue virus type 1 and type 3 as dominant serotypes during 2017 in Pune and Nashik regions of Maharashtra, Western India. Infect Genet Evol. 2018;66:272-283.
    https://doi.org10.1016/j.meegid.2018.10.016
17. Alagarasu K, Patil JA, Kakade MB, More AM, Yogesh B, Newase P, et al. Serotype and genotype diversity of dengue viruses circulating in India: A multi-centre retrospective study involving the Virus Research Diagnostic Laboratory Network in 2018. Int J Infect Dis. 2021;111:242-252.
    https://doi.org/10.1016/j.ijid.2021.08.045
18. Racherla RG, Pamireddy ML, Mohan A, Mudhigeti N, Mahalakshmi PA, Nallapireddy U, et al. Co-circulation of four dengue serotypes at South Eastern Andhra Pradesh, India: A prospective study. Indian J Med Microbiol. 2018;36(2):236-240.
    https://doi.org/10.4103/ijmm.IJMM_18_109

19. Singh PS and Chaturvedi HK. Temporal variation and geospatial clustering of dengue in Delhi, India 2015-2018. BMJ Open. 2021;11(2):e043848.
https://doi.org/10.1136/bmjopen-2020-043848
20. Debnath F, Provash CS, Chakraborty A and Dutta S. Dengue fever outbreak by more than one serotype in a municipal area of Kolkata, Eastern India. J Vector Borne Dis. 2019;56(4):380-382.
https://doi.org10.4103/0972-9062.302043
21. Endy TP, Nisalak A, Chunsuttiwat S, Libraty DH, Green S, Rothman AL, et al. Spatial and temporal circulation of dengue virus serotypes: Aprospective study of primary school children in Kamphaeng Phet, Thailand. Am J Epidemiol. 2002;156(1):52-59.
https://doi.org/10.1093/aje/kwf006
22. Kalayanarooj S and Nimmannitya S. Clinical and laboratory presentations of dengue patients with different serotypes. WHO Dengue Bull. 2000;24:53-9.
23. Vaughn DW, Green S, Kalayanarooj S, Innis BL, Nimmannitya S, Suntayakorn S, et al. Dengue viremia titer, antibody response pattern, and virus serotype correlate with disease severity. J Infect Dis. 2000;181:2-9.
https://doi.org/10.1086/315215
24. Nisalak A, Endy TP, Nimmannitya S, Kalayanarooj S, Thisayakorn U, Scott RM, et al. Serotype-specific dengue virus circulation and dengue disease in Bangkok, Thailand from 1973 to 1999. Am J Trop Med Hyg. 2003;68(2):191-202.
25. Yung CF, Lee KS, Thein TL, Tan LK, Gan VC, Wong JG, et al. Dengue serotype-specific differences in clinical manifestation, laboratory parameters and risk of severe disease in adults, Singapore. Am J Trop Med Hyg. 2015;92(5):999-1005.
https://doi.org/10.4269/ajtmh.14-0628

Authors' Contributions:
SB- Definition of intellectual content, literature survey, prepared first draft of the manuscript, implementation of the study protocol, data collection, data analysis; **FDL**- Design of study, statistical analysis, and interpretation; **PPM**- Literature survey, preparation of tables and figures, manuscript revision; **AB**- Concept, design, clinical protocol, manuscript preparation, editing, and manuscript revision, manuscript review.

Work attributed to:
Department of Microbiology, Tamralipto Government Medical College and Hospital, Purba Midnapore, West Bengal, India.

Orcid ID:
Dr. Sayani Bose - https://orcid.org/0009-0006-3578-4732
Dr. Feradica D Laloo - https://orcid.org/0009-0003-8425-8219
Dr. Partha Pratim Maity - https://orcid.org/0009-0006-3257-6946
Dr. Anirban Bhaduri - https://orcid.org/0000-0001-6131-4597

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