

# Infection Epidemiology of Dengue Virus and Its Impacts on Liver Enzymes

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DOI: <https://doi.org/10.36348/sjpm.2024.v09i09.003>

| Received: 20.08.2024 | Accepted: 26.09.2024 | Published: 28.09.2024

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## Abstract

**Background:** Dengue fever is a mosquito-borne viral disease that has emerged as a significant public health concern in Kolkata, with a substantial burden on the healthcare system. Also, this disease shows an additional impact on liver. **Objective:** To investigate the epidemiological trends of dengue fever in Kolkata and its effect on liver function. **Methods:** A retrospective study was conducted using data from [Ashok Laboratory] in Kolkata. Patients diagnosed with dengue fever between [July, 2023 to June, 2024] were included. Demographic, clinical, and laboratory data were analyzed. **Results:** A total of [6132] patients were included. The majority were [10-30] years old, and [males]. The incidence of dengue fever was highest during [monsoon season]. The data shows that SGPT/ALT and SGOT/AST value is very high on the dengue positive individuals and total bilirubin value is also high on dengue positive individuals. **Conclusion:** This study highlights the significant impact of dengue fever on liver function in Kolkata. The findings suggest that early detection and management of dengue-induced hepatic dysfunction are crucial to prevent severe complications. Public health measures should focus on vector control, timely diagnosis, and appropriate treatment to mitigate the burden of dengue fever in Kolkata.

**Keywords:** Dengue fever, mosquito-borne viral disease, epidemiological trends, SGPT/ALT, SGOT/AST, hepatic dysfunction.

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## INTRODUCTION

Dengue is the most rapidly spreading mosquito-borne viral disease in the world [1]. Dengue is a vector-borne viral disease caused by the flavivirus dengue virus (DENV). Approximately 400 million cases and 22 000 deaths occur due to dengue worldwide each year [2]. Dengue is an acute viral illness caused by RNA virus of the family Flaviviridae and spread by Aedes mosquitoes [3]. The dengue virus, a member of the genus Flavivirus of the family Flaviviridae, is an arthropod-borne virus that includes four different serotypes (DEN-1, DEN-2, DEN-3, and DEN-4) [4]. Dengue virus is transmitted through the bite of the Aedes mosquito and has been observed to have hepatotoxic effects. Liver involvement is common, leading to abnormalities in liver function tests, including mild elevations in serum bilirubin, increased transaminases, and alterations in serum albumin. While most cases are asymptomatic, liver dysfunction may occasionally manifest clinically as jaundice or, in severe cases, acute liver failure (ALF).

This highlights the importance of monitoring liver function in dengue patients to prevent and manage potential hepatic complications [5].

Aim of the study is to investigate the epidemiological trends of dengue fever in Kolkata, including incidence, prevalence, and seasonality and to examine the relationship between dengue infection and liver dysfunction in patients from Kolkata.

Objectives of the study is to analyze the demographic characteristics of dengue patients in Kolkata, to determine the frequency and severity of liver dysfunction in dengue patients from Kolkata, the most effective season of dengue virus in Kolkata and to determine the age group which is mostly affected by the dengue virus.

## METHODOLOGY

Test samples were collected randomly from every zone of Kolkata, west Bengal. The study was

studied from July, 2023 to June, 2024. It is a one-year study where all total 6132 samples were studied for the dengue test. 4818 blood samples were taken at the onset of fever and allowed to NS1 dengue test while 893&421 samples were taken after four or five days of the fever and allowed to IgM and IgG antibody test respectively.

Among those 6132 test samples 39 dengue positive test samples and 39 dengue negative test samples were taken randomly and allowed to lipid profile test.

The whole test procedure was performed in the Ashok Laboratory, Kolkata. NS1, IgG, IgM tests were performed by using NS1, IgG, IgM micro-Elisa kit, and lipid profile test were performed by using COBAS 6000 (c501) machine, based on the principle of ion selective electrode (ISE) and photometric analysis.

**The test procedure for NS1:** To 50  $\mu$ l sample diluent, 50  $\mu$ l serum was added. 50  $\mu$ l diluent and 50  $\mu$ l negative control was placed in the another well, about 50  $\mu$ l diluent and 50  $\mu$ l calibrator were taken on three different wells and 50  $\mu$ l diluent and 50  $\mu$ l positive control was taken one another well. Then 100  $\mu$ l enzyme conjugate were added on each well, which was incubate at 37°C for 90 minutes. This all mixture were discarded and the wells were washed with wash buffer (20ml prepared 25X wash buffer concentrate with 480ml distilled water) for six times. Then to the wells 150  $\mu$ l of working substrate was added and kept for 30 minutes at room temperature in dark place. After that 100  $\mu$ l stop solution were added on each well and the sample was read in ELISA reader at 450/630 nm. Cut-off point was determined ( $\geq 1.5$ \*Negative Control O.D). Followed by the kit procedure of J. Mitra.

**The test procedure for IgG and IgM:** To 1 ml sample diluent, 10  $\mu$ l serum was added to dilute the sample and from this diluted sample 100  $\mu$ l diluted sample was taken. 100  $\mu$ l negative control was placed in the another well, about 100  $\mu$ l calibrator were taken on three different wells and 100  $\mu$ l positive control was taken one

another well. Then allowed to incubate at 37°C for 60 min. This all mixture were discarded and the wells were washed with wash buffer (20ml prepared 25X wash buffer concentrate with 480ml distilled water) for five times. Then to the wells 100  $\mu$ l of enzyme conjugate was added and incubated at 37°C for 60 min. Then again, this all mixture were discarded and the wells were washed with wash buffer (20ml prepared 25X wash buffer concentrate with 480ml distilled water) for five times. Then 100  $\mu$ l working substrate was added in each well and kept for 30 minutes in room temperature at a dark place. After that 50  $\mu$ l stop solution were added on each well and the sample was read in ELISA reader at 450/630 nm. The procedure was same for IgG and IgM antibodies. Cut-off point was determined ( $\geq 1.5$ \*Negative Control O.D). Followed by the kit procedure of J. Mitra.

**The test procedure for Lipid Profile Test:** This test is performed in COBAS (c501) machine, which is a software-controlled machinery test, where all reagents are store inside the machine and the machine uses the reagents according to the needs of the test which will perform. It utilizes spectrophotometric and ion-specific electrode measuring system to perform the test. Here only serum sample is to be inserted inside the machine and then rest of the test is performed by the machine independently. Followed by the kit procedure of Roche.

## RESULT

### YEARLY (July, 2023 – June, 2024) DISTRIBUTION OF DENGUE

One year (July, 2023 – June, 2024) dengue positive cases were studied and total 1581 were tested positive from random test samples in Kolkata, West Bengal. Separately data were taken in each month and prepared a yearly distribution graph. The data shows a high surge on august (271), September (662) and October (494) month among positive test samples. The cases are rising with a higher rate from August and getting decreased from the October month. Majorly it is observed that the rate of dengue is especially high in monsoon season.

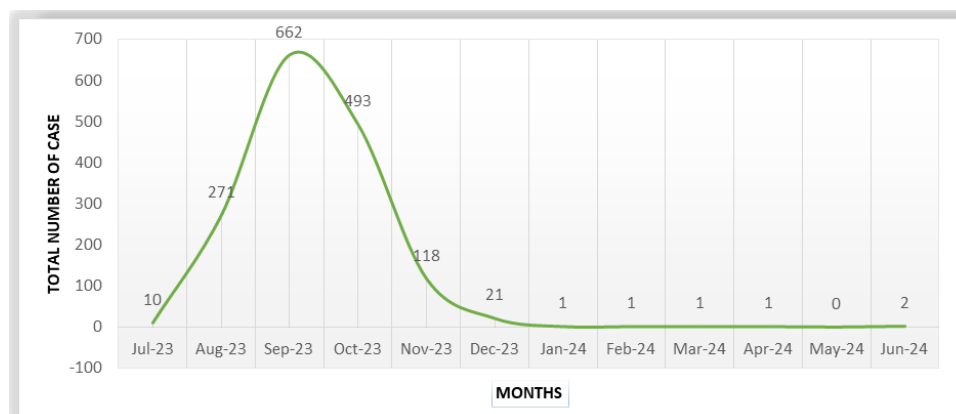
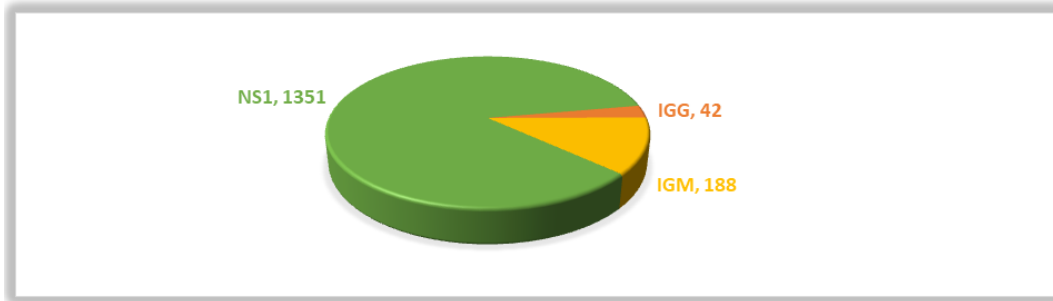


Figure 1: Yearly Distribution Graph of Dengue Positive Cases

**DENGUE POSITIVE CASES DISTRIBUTION BASED ON ANTIBODIES AND ANTIGENS PRESENT IN BLOOD**

Among 1581 positive test samples, two types of tests were performed. One for antibodies (IGG, IGM)

and another for antigens (NS1). We prepared a pie chart on the basis of IGG, IGM antibodies and NS1 antigens found, where 1351 individuals were tested NS1 positive, 42 were tested IgG positive and 188 were tested IgM positive.

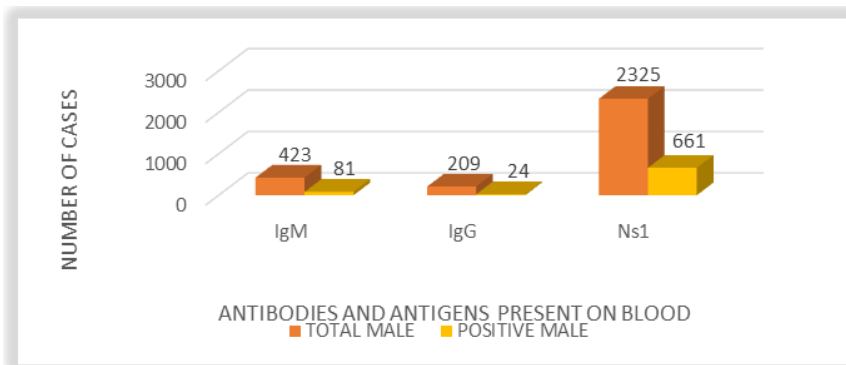


**Figure 2: Dengue Positive Cases Distribution Based on Antibodies and Antigens**

**TOTAL DENGUE TEST SAMPLE AND POSITIVE MALE, POSITIVE FEMALE DENGUE TEST SAMPLE DISTRIBUTION**

Total 6132 test samples were taken for the study; among them 1581 test samples were tested positive. We segregate the data into males and females. total 2957 random male test samples were taken for the

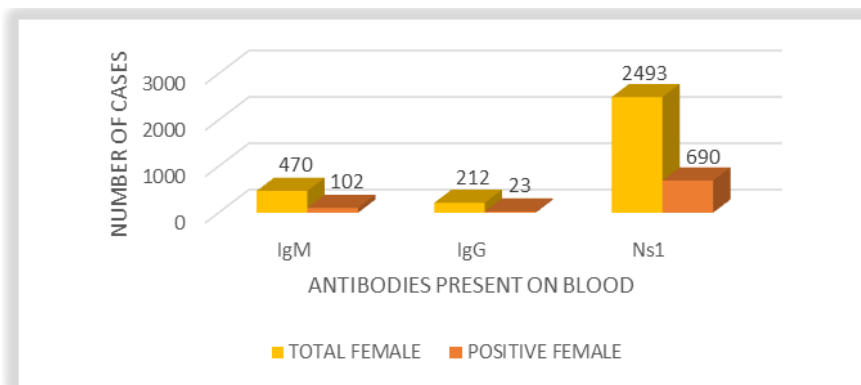
test and 766 males were tested positive. The rate of positive male is 25.90%. Among 766 positive cases 661 males were tested NS1 positive out of 2325 random tested samples, 81 males were tested IgM positive among 423 random test samples and 24 males were tested IgG positive among 209 test samples.



**Figure 3: Dengue Year Wise Total Male Test Samples and Positive Male Test Samples Distribution**

Total 3175 random female test samples were taken for the test and 815 females were tested positive. The rate of positive female is 25.60%. Among 815 positive cases 690 females were tested NS1 positive out

of 2493 random tested samples, 102 females were tested IgM positive among 470 random test samples and 23 females were tested IgG positive among 212 test samples.

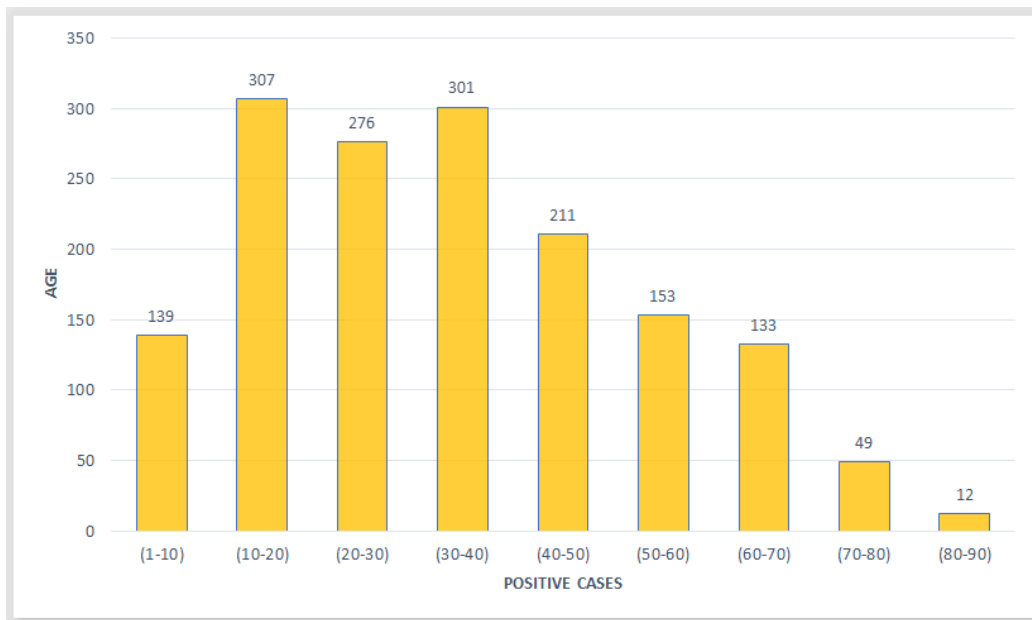


**Figure 4: Dengue Year Wise Total Female Test Samples and Positive Female Test Samples Distribution**

From the above data we studied that male shows higher dengue positive rates than females. It also suggests that NS1 antigens shows higher rate of infection both for male and female.

**AGE WISE DENGUE POSITIVE CASE DISTRIBUTION BASED ON AGE –**

We also studied the data based on different age groups. Total 1581 positive test samples were segregated into different age groups and then prepared a bar graph of the study.



**Figure 5: Age Wise Dengue Positive Case Distribution Based on Age**

The above data suggests that 10-20, 20-30, 30-40 age groups are mostly affected by the dengue virus. While 10-20 age group shows the high number of affected individuals.

**EFFECTS OF DENGUE VIRUS ON LIVER**

Among 1581 positive test samples 39 samples were randomly taken and studied their liver function test and also 39 random dengue negative samples were taken and studied their liver function test. Then those samples were accumulated to perform pair t-test. In liver function test total protein, total bilirubin, SGPT/ALT, SGOT/AST and alkaline phosphatase were studied. P-value for total protein is ~0.06, P-value for total bilirubin

is ~0.21, P-value for SGPT/ALT is ~0.0008, P-value for SGOT/AST is ~0.00001 and P-value for alkaline phosphate is ~0.003. This study suggests that values of SGPT/ALT, SGOT/AST and alkaline phosphate matches the confidence interval of P-value (SGOT/AST, SGPT/ALT 99.9% confidence interval and Alkaline phosphate 99% confidence interval) and hence the values are statistically significant. While total protein and total bilirubin's P-value suggests they are statistically insignificant. From the data of 39 dengue infected and 39 dengue non-infected sample we prepared a mean with +/- 2 standard division of liver function test which is given below.

**Table 1: Statistical Calculation of Liver Function Test of Dengue Positive Compared with Normal Individuals**

Test Performed	Dengue Infected (Mean+/- 2SD)	Dengue Non-Infected (Mean+/- 2SD)	P-Value
TOTAL PROTEIN	8.27	8.45	~0.06
TOTAL BILIRUBIN	1.30	1.14	~0.021
SGPT/ALT	209.19	71.70	~0.0008**
SGOT/AST	243.90	58.08	~0.00001**
Alkaline Phosphate	207.71	106.33	~0.003*

\*: Statistically significant with 99% confidence interval (p-value - < 0.01)  
 \*\*: Statistically significant with 99.9% confidence interval (p-value - < 0.001)

**DISCUSSION**

Dengue is one of the major public health threats in Kolkata [6]. The first virologically proved epidemic of dengue fever (DF) occurred in Kolkata and Eastern Coast of India in 1963-1964 [7, 8]. This disease affects

liver functions with an imbalance of liver enzymes followed by other clinical manifestations [9].

From the data that we collected we can categorize the dengue results in several distinct group

SGPT/ALT and SGOT/AST value is very high on the dengue positive individuals and total bilirubin value is also high on dengue positive individuals., that means the effectiveness of dengue only remains active on the monsoon season in Kolkata, West-Bengal. Another study of some researchers also concluded on the same topic [10]. West Bengal's dengue case count for 2022 was the highest countrywide and the evidence says that temperature, humidity and rainfall are key climatic factors for the breeding of Aedes mosquitoes. The late withdrawal of monsoon and warming global temperatures have stretched the dengue transmission window in India [11].

Another analysis says that 25.90% males were tested dengue positive and 25.60% females were tested dengue positive. Although the ratio is almost same but the rate of dengue is slightly higher in males than females. Another evidence suggests that the ratio of male-infected individuals was higher (18,281, n = 25,682) than female individuals (7,301, n = 25,682) [12].

Another analysis of dengue positive data was done on the basis of age group, where it is clearly observed that 10-40 age group's people are mostly affected by the dengue virus. It's may be due the high exposure on outside of that age grouped peoples. Another study also suggests the same [13].

We take random 39 dengue positive individuals and 39 dengue negative individuals to perform lipid profile test. The data shows that SGPT/ALT and SGOT/AST value is very high on the dengue positive individuals and total bilirubin value is also high on dengue positive individuals. This data suggests that dengue virus also causes effect and damage the liver cells. Another study of some researchers suggests that association between viral load and liver enzymes shows a positive correlation with an increased level of T.BIL, ALT, and AST, while all other parameters also show a significant association with a p-value of less than 0.0001 [14].

## ACKNOWLEDGEMENT

I would like to express my sincere gratitude to Diganta Dey for their guidance, support, and expertise throughout this research project. Their valuable insights and constructive feedback have been instrumental in shaping this study.

I would also like to thank Ashok Laboratory for providing the necessary resources and facilities to conduct this research.

Furthermore, I would like to acknowledge the contributions of Mithun Das, Swati Dalui, Susmita Sett, Birinchi Adhikary, Chittaranjan Maity, Rahul Ghosh, Kingkar Ballav, Joydip Sasmal, Biswajit barman, Mita

majumder, Ranu mondal, Saibal mandal who assisted with data collection, data analysis, and manuscript preparation.

Finally, I would like to thank my family and friends and teachers for their unwavering support and encouragement throughout this research journey.

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