

Association of Demographic Profiles, Clinical Features and Laboratory Parameters in Seropositive Dengue Virus Infection at a Tertiary Care Hospital: A Cross Sectional Study

Research Article

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Abstract

Background: Dengue virus infection remains a significant global health concern, particularly in regions with tropical and subtropical climates. As one of the most prevalent mosquito-transmitted diseases, Dengue fever affects over 128 countries worldwide. Bangladesh, in particular, has experienced recurrent outbreaks of Dengue since the year 2000, with the situation escalating to a critical level in 2019, marking it as one of the most severe outbreaks in the country's recent history. The clinical and laboratory features of Dengue virus infection are crucial for effective management and intervention.

Objective: To comprehensively examine the clinical and laboratory aspects of Dengue virus infection in patients admitted to a Tertiary Care Hospital.

Material and Methods: This cross-sectional study took place between June 2023 to December 2023 in Bashundhara Ad-din Medical College & Hospital, South Keranigonj, Dhaka, Bangladesh. Data pertaining to demographic details, clinical attributes, and laboratory profiles of 271 confirmed cases of hospitalized acute dengue were gathered through the utilization of a structured questionnaire. Subsequent to data collection, thorough cleaning and analysis were conducted using the statistical software SPSS version 25.

Results: This study showed the mean age of the participants was 26.21 years, with nearly half of them (48.8%) falling within the 20–40 years age bracket. Predominant clinical symptoms included fever (93.7%), abdominal pain (29.9%), skin rash (25.5%), and diarrhea (19.9%). Complications were observed in 159 patients, with prevalent issues such as breathing problems (41.7%), pleural effusion (38.4%), and gum bleeding (11.4%). A substantial majority, exceeding 90%, exhibited seropositivity for the DENV-NS1 antigen.

Conclusions: The study highlighted the severity of Dengue, with a substantial number of hospitalizations and confirmed deaths. Early diagnosis, especially through NS1 antigen detection, played a crucial role. We identified risk factors for severe Dengue and observed diverse clinical presentations. The study sheds light on the demographic profile and emphasizes the need for continued research and public health efforts to manage Dengue effectively.

Keywords: Dengue, Pleural Effusion, NS1 Antigen

Introduction

“Dengue fever,” also known as “Break-bone fever,” is the most prevalent acute systemic viral infection transmitted by arthropods [1-3].

Dengue virus infection (DENV) stands as the most common mosquito-borne infectious ailment on a global scale. Considered endemic in over 100 countries, dengue affects approximately 2.5 billion people residing in dengue-prone regions, with an annual reporting of around



100 million new cases [4]. The primary vector responsible for transmitting the dengue virus is the *Aedes aegypti* mosquito. The pathogenic female *Aedes* mosquito conveys the virus to humans through bites, often acquiring the virus while feeding on the blood of an infected person. These mosquitoes thrive in stagnant water, such as water tanks, puddles, old tires, and containers [1-5].

Over the last five decades, the occurrence of dengue has surged by 30 times, and Bangladesh bears one of the highest dengue burdens globally [5]. Since the year 2000, Bangladesh has experienced a dengue outbreak nearly every year, with over 3000 reported cases in at least six of these yearly outbreaks [6]. In 2019, the country witnessed a significant impact, with more than 100,000 individuals hospitalized due to Dengue Virus (DENV) infection. Notably, around 50% of these cases were concentrated in Dhaka City, the capital of Bangladesh. According to the Directorate General of Health Services (DGHS), there were 164 confirmed deaths attributed to dengue in 2019 [5-7]. The manifestations of dengue range from non-specific febrile symptoms to classic dengue fever, often accompanied by hemorrhage or shock (Dengue Shock Syndrome) [7].

Initially, common clinical indicators include fever, nausea, vomiting, skin rash, and body aches. Classic dengue fever is characterized by a sudden onset of high fever (up to 40°C), intense headache, nausea, vomiting, severe joint and muscle pain, retro-orbital pain, and a spreading maculopapular rash [5]. In contrast, severe dengue symptoms emerge 1–2 days after the fever subsides and include tenderness, abdominal pain, frequent vomiting, nose bleeding, vomiting of blood (hematemesis), dark stools (melena), fatigue, and restlessness [1-5]. While most dengue cases resolve on their own, untreated cases can progress to a life-threatening condition. The onset of severe dengue can be swift, leading to internal hemorrhage, organ failure, a drastic drop in blood pressure resulting in shock, and complications such as pre-term birth and very low birth weight during pregnancy [2-5].

Risk factors for severe dengue include secondary dengue infection, elderly patients, elevated hematocrit levels, low platelet count, and prolonged activated partial thromboplastin time (APTT). Emergency hospitalization is imperative for patients exhibiting these parameters [5-8]. The rapid onset of severe dengue contributes to numerous deaths among children and young adults, particularly in Asian and Latin American countries [4,5]. Early detection is essential for preventing fatalities resulting from dengue infection. Differential and diagnostic confirmation typically involve routine laboratory tests such as complete blood count (CBC), blood culture, or serological examination. The primary method for early dengue diagnosis is detecting the NS1 antigen in the blood, as the IgM antibody becomes detectable only after the 6th day of clinical manifestation [9]. Nevertheless, clinical insights obtained from patient history, physical examinations, and routine laboratory tests remain pertinent in diagnosing dengue cases [5-9].

The blood profile of individuals with dengue undergoes changes from the onset of fever. Typically, thrombocytopenia develops within 3–8 days, followed by leukopenia and hemoconcentration due to plasma leakage [3-5]. Statistics indicate an 87% increase in leukopenia and a positive tourniquet test in 52% of dengue patients. Consequently, individuals presenting with acute febrile leukopenia and a positive tourniquet test are more likely to receive a diagnosis of dengue rather than influenza, enteroviruses, or leptospirosis [5-8]. A precise understanding of the clinical and laboratory profile is crucial for accurate diagnosis and effective management of patients. Therefore, this study aims to elucidate the clinical and laboratory profile of serologically confirmed dengue cases in Bangladesh.

Materials and Methods

This prospective observational study was conducted between June 2023 and December 2023 in Dhaka city, which is the primary area of the dengue outbreak, served as the focal point. The research took

place in the Bashundhara Ad-din Medical College & hospital, South Keranigonj, Dhaka. A total of 271 dengue patients admitted to the hospital from June to December 2023 were included in the study. All patients had confirmed dengue based on the positivity of NS1 (non-structural protein) antigen. Thorough monitoring of admitted patients was conducted, with regular recording of essential clinical and laboratory details on a standard case report form. Comprehensive clinical examinations, encompassing vital signs, skin rashes, pleural effusion, breathlessness, ascites, hepatomegaly, and splenomegaly, were diligently performed. Patients were included based on laboratory confirmation of NS1 Ag or Anti-dengue IgM, presenting with an oral temperature of 100.4 °F, <7 days of fever, and reporting at least one specific symptom (headache, joint pain, backache, abdominal pain, vomiting, fatigue, anorexia, or diarrhea). Recruitment was irrespective of age, gender, economic class, or ethnicity. Informed consent was obtained from non-critically ill patients, while guardians provided assent for critically ill patients. Patients with diagnoses other than dengue were excluded from the study.

Sample collection and processing involved obtaining 3 mL of venous blood through venipuncture from each patient. Subsequently, the blood was centrifuged, and plasma was transferred into EDTA tubes. Plasma aliquots were prepared and stored at -20 °C for future analysis. NS1 antigen detection was performed on each patient's plasma. The Tell me fast® Combo Dengue NS1-IgG/IgM Rapid Test (Biocan Diagnostics Inc. Canada) was used to identify IgM antibodies, and the analysis followed the manufacturer's guidelines. To validate IgM and IgG antibodies against the dengue virus, an indirect enzyme-linked immunosorbent assay (ELISA: EUROIMMUN diagnostics) was employed. Confirmed acute dengue cases were defined as patients with samples positive for DENV NS1 protein alone or DENV NS1 protein with IgM antibodies or DENV NS1 protein with IgG antibodies against DENV, exhibiting febrile illness, and reporting at least one of the specified symptoms: headache, backache, abdominal pain, joint pain, vomiting, anorexia, fatigue, or diarrhea. Routine hematological laboratory investigations, including complete blood cell count (CBC) and hematocrit level, were analyzed using an automated blood analyzer. Biochemical tests such as aspartate aminotransferase (AST), alanine transaminase (ALT) for liver function tests, creatinine level, etc., were conducted using an automated biochemistry analyzer (Vegasys). All patients underwent clinical examination by a registered physician, and clinical features, as well as laboratory parameters, were recorded by registered nurses using a structured questionnaire. Cutoff values for each investigation were based on reference ranges provided by the laboratory. The accuracy and completeness of the data were thoroughly checked, and data cleaning and analyses were performed using statistical software SPSS version 25.

Results

In this investigation, a total of 271 participants were enrolled, all of whom tested positive for the seroprevalence of acute dengue virus (DENV) infection. Among these individuals, 80.8% were in the recovery phase, and 129.2% were in the critical phase (Table 1). Of the total, 60.1% were male, and 39.9% were female. The mean age of all participants was 26.21 (±14.99) years, with almost half (49.8%) falling within the 20–40 age range. Children (<10 years) comprised 12.5% of the subjects, while adolescents (10–19 years) made up 22.5%. Only 15.1% were over 40 years old. Regarding family structure, 65.7% had a nuclear family, and the mean number of family members was 4.82 (±2.65), with 44.6% having more than four family members. Approximately half (50.6%) of the patients were unemployed, and the majority resided in urban areas (69.7%), with the remaining 30.3% hailing from semi-urban or rural areas (Table 2).

Fever was reported by 93.7% (254) of the patients, with myalgia, maculopapular skin rash, and itchy skin noted in 26.6%, 25.5%, and 21.8% of the cases, respectively. Abdominal pain and diarrhea were reported by 29.9% and 19.9%, respectively. Nausea and vomiting were



experienced by 61.6%, while headaches affected 45.8% of the patients. Less common symptoms included retro-orbital pain in 4.8% and conjunctival suffusion in 2.2%. Additionally, 6.6% reported other complaints such as anorexia, fatigue, and epistaxis (Table 3). Among the 271 patients, 58.7% presented with dengue complications. Hemorrhagic manifestations were found in 11.4%, pleural effusion in 38.4%, and breathing problems in 41.7%. Ascitic fluid exudation was documented in 29.9%, while hepatomegaly and splenomegaly were registered in 7.7%. Multiple organ failure was reported in 9.6% of cases (Table 4).

common abnormality, affecting 73.4% of patients. Among these, 32.5% also had leukopenia (leukocyte count <4000/cumm). Elevated liver enzyme levels (AST, ALT >45 IU/L) were identified in 51.3% of patients. A marked increase in hematocrit level (>45%) was observed in 55.2% of participants (Table 5,6). Regarding seropositivity, 93.7% (254 out of 271) tested positive for the DENV-NS1 antigen alone, while 92.6% (251 out of 271) exhibited dual seropositivity to DENV-NS1 plus anti-DENV IgM. Only 6.6% showed dual seropositivity to DENV-NS1 plus anti-DENV IgG antibodies. The tourniquet test, a clinical diagnostic method, yielded a positive result for DENV infection in 17.7% of the enrolled patients (Table 7).

Thrombocytopenia (platelet count <50,000/cumm) was the most

Table 1: Health status of the enrolled patients with DENV infection (n=271).

Health Status	Number of Patients	Percentage (%)
Recovery phase	219	80.8
Critical phase	52	19.2

Table 2: Demographic characteristics of the patients (n=271).

Characteristics	Number of Patients	Percentage (%)
Gender		
Male	163	60.1
Female	108	39.9
Age (in years)		
<10	34	12.5
10–19	61	22.5
20–40	135	49.8
>40	41	15.1
Mean±SD	26.21 ± 14.99	
Family Type		
Nuclear	178	65.7
Joint	93	34.3
Number of family members		
4	121	44.6
5	68	25.1
>6	82	30.3
Mean ± SD	4.82 ± 2.65	
Occupation		
Employed	137	50.6
Unemployed	134	49.4
Residence		
Semi-Urban/Rural	82	30.3
Urban	189	69.7

Table 3: Clinical Features of patients (n=271).

Clinical Features	Number of Patients	Percentage (%)
Fever	254	93.7
Abdominal Pain	81	29.9
Diarrhea	54	19.9
Skin Rash	69	25.5
Itching	59	21.8
Myalgia	72	26.6



Nausea/Vomiting	167	61.6
Headache	124	45.8
Conjunctival suffusion	6	2.2
Retro-Orbital Pain	13	4.8
Others (Anorexia, Fatigue and Epistaxis)	18	6.6

Table 4: Complications of the patients (n=271). Total will not correspond to 100% because of multiple complication in individual patients.

Complications	Number of patients	Percentage (%)
None	112	41.3
Bleeding	31	11.4
Pleural Effusion	104	38.4
Breathlessness	113	41.7
Ascites	81	29.9
Hepatomegaly	21	7.7
Splenomegaly	3	1.1
Seizures	9	3.3
Multiple Organ Failure	26	9.6

Table 5: Distribution of patients with complications according to lab parameters (n=271).

Complications Due To DENV Infection	Platelet Count		Leukocyte Count		Liver Enzyme		Hematocrit	
	≤50,000	>50000	≤4000	>4000	Raised AST, ALT	Normal AST, ALT	Normal	Raised
Bleeding (n=31)	13	18	14	17	18	13	19	12
Pleural Effusion (n=104)	39	65	35	69	65	39	59	45
Ascites (n=81)	25	56	35	46	46	35	48	33
Breathlessness (n=113)	24	89	38	75	65	48	57	56
Seizures (n=9)	2	7	6	3	1	8	6	3
Hepatomegaly (n=21)	9	12	6	15	16	5	8	13
Multiple Organ Failure (n=26)	4	22	13	13	13	13	9	17

Table 6: Laboratory parameters of the patients (n=271).

Laboratory Parameters	Number of Patients	Percentage (%)
Leukocyte Count (N=209)		
≤4000/cumm	68	32.5
>4000/cumm	141	67.5
Platelet Count (n= 271)		
<50000/cumm	199	73.4
≥50000/cumm	72	26.6
Liver Enzyme (n =271)		
Raised AST, ALT (>45 IU/L)	139	51.3



Normal AST, ALT	132	48.7
Hematocrit Value (n=252)		
Raised Hematocrit (>45%)	139	55.2
Normal Hematocrit	113	44.8

Table 7: Pattern of seropositivity and clinical diagnostic method of patients (n=271).

Lab Parameters	Number of Patients	Percentage
Ns1		
Positive	254	93.7
Negative	17	6.3
Igm		
Positive	251	92.6
Negative	20	7.4
Igg		
Positive	18	6.6
Negative	253	93.4
Tourniquet Test		
Positive	48	17.7
Negative	223	82.3

Discussion

In the past few years, dengue has demonstrated a dynamic surge and has emerged as a significant global challenge. The escalation in dengue cases can be attributed to the escalating, unplanned urbanization characterized by unregulated infrastructure development and inadequate sanitation facilities, creating numerous breeding grounds for mosquitoes. In Bangladesh, the majority of dengue cases were documented during the monsoon season (50%) and the post-monsoon period (49%), with the peak season occurring from July to October [10]. Similar to other Southeast Asian nations, Bangladesh, situated in tropical and sub-tropical zones, has become an optimal environment for the dengue vector, fostering its increased transmission. Both *Aedes aegypti* and *Aedes albopictus*, the vector types, were identified during dengue outbreaks from 2000 to 2017 [11].

Examining the dengue scenario in Southeast Asia provides an insight into the escalating economic and social challenges posed by this emerging disease, with a pronounced impact in Southeast Asia, particularly Bangladesh. Analogous to other low- and middle-income countries (LMICs), the current state of dengue in Bangladesh is exerting economic pressures on the healthcare sector, as evidenced by a consistent year-on-year decline in healthcare expenditure allocation [10]. Concurrently, out-of-pocket expenditure (OOP) is on the rise, reaching 67%, the highest in the South-East Asia region, as indicated by findings from the Bangladesh National Health Accounts study (BNHA-V) [12]. Dengue transmission reaches its zenith during the rainy season, specifically from August to October, creating optimal conditions for the *Aedes aegypti* mosquito [13].

In this current investigation, the analysis revealed a higher incidence of dengue fever in men compared to women, aligning with findings from prior studies in Saudi Arabia [14] and Nepal [15], although differing from a study in Cameroon [16]. The disparity between males and females may be attributed to the increased exposure of men to mosquitoes carrying the virus, either in the workplace or during commuting. The majority of dengue cases (49.8%) in this study occurred within the 20–40 age group. This pattern is consistent with El-Gilany’s discovery of a higher prevalence among individuals aged 16–44 in Saudi Arabia [14] and M. Rahman et al. report of the highest proportion of cases among the 18–33 age group in Bangladesh [17] Both

studies indicate a greater occurrence in adults, aligning with our findings.

Observations also revealed a lower impact of dengue fever on young children under ten years old (12.7%). Similar trends were documented in Nepal [13], Nigeria [18], and Cameroon [19]. The reduced prevalence of dengue infection in children compared to the elderly might be explained by the additional care provided to children by their parents. Dengue diagnosis necessitates either the direct identification of the virus or the detection of specific antibodies. Swift diagnosis is crucial for promptly treating patients. While the conventional “gold standard” involves the isolation and identification of the virus, the real-time reverse transcriptase-polymerase chain reaction (RT-PCR) method is gradually superseding it due to its rapid diagnostic capabilities [19]. However, owing to its affordability and ease of implementation in developing nations, the enzyme-linked immunosorbent assay (ELISA) method for detecting NS1 antigen or specific IgG and IgM antibodies (individually or in combination) remains a vital diagnostic tool compared to RT-PCR [20].

Combining DENV (NS1) with specific antibodies (IgM and IgG) offers an additional advantage by enhancing the accuracy of dengue diagnosis and mitigating the risk of false-positive results from a single test [21,22]. Hunsperger et al [23]. reported sensitivity and specificity values for the NS1 antigen method ranging from 60–75% and 71–80%, respectively. In the case of IgM anti-DENV ELISA, the range was 96–98% and 78–91%, respectively. Another study by V. Tricou et al [20]. demonstrated that including IgM/IgG test results significantly increased the sensitivity of NS1 alone, from 62.4% to 75.5% when NS1 and/or IgM tested positive, and 83.7% when NS1 and/or IgM and/or IgG were positive.

In our present study, both single and combined methods were employed on 271 serum samples from febrile patients. We observed a higher detection rate of Dengue cases in the NS1 plus IgM antibodies test and NS1 antigen test alone compared to the tourniquet test and NS1 plus IgG antibodies test. These findings align with a study conducted by C. Palomares-Reyes et al. in Peru but differ from those reported by O. G. Oyero in Nigeria and A. M. Ashshi et al. in Saudi Arabia [24,25].



The clinical presentation of dengue patients included in the present survey indicates that fever was the predominant symptom (93.7%), aligning with findings from studies in Bangladesh, [5] Pakistan [26], Saudi Arabia [14], and India [27]. Additionally, patients exhibited symptoms such as nausea-vomiting, headache, abdominal pain, myalgia, and skin rash. Badreddine et al [28] highlighted abdominal pain and vomiting as more prevalent symptoms in their research. Another study by Abdel-Hady El-Gilany [14] identified headache (74.60%) and myalgia (67.60%) as the most common symptoms after fever, indicating a higher percentage than observed in our study.

Skin rash was observed in 25.5% of dengue patients, consistent with earlier studies by Mahmood et al [5], El-Gilany [14] in Bangladesh and Saudi Arabia, and Ramabhatta 29 in India. Ocular manifestations, such as conjunctival suffusion and retro-orbital pain, were less frequently reported in this study compared to other investigations [14-29]. Notably, among the 271 participants in this study, 17 patients did not exhibit fever during the data collection session, with 16 of them reporting having had a fever either on the previous day or a few hours earlier. This discrepancy might be attributed to the use of antipyretic drugs by the patients.

In the present study, breathlessness (41.7%) emerged as the most prevalent complication of dengue, followed by pleural effusion (38.4%) and ascites (29.9%). These findings are consistent with a prior study conducted in Bangladesh [5]. Godbole [30] reported pleural effusion and ascites in 31% of dengue patients in India, which is comparatively lower than the rates observed in our study. Bleeding was observed in 11.4% of patients, indicating a higher incidence compared to the current study findings (6.5%). Several studies have highlighted both typical and atypical complications of dengue fever, encompassing acute respiratory distress syndrome (ARDS), dengue encephalopathy, encephalitis, lymphadenopathy, splenomegaly, myocarditis, anemia, multiple organ failure, hepatitis, febrile diarrhea, refractory shock, impaired consciousness, portal hypertension, appendicitis, pericardial effusion [31-33].

Following the examination of laboratory investigations in our study, the results indicate that thrombocytopenia was the most prevalent hematological abnormality among the patients, affecting 73.4% of them. This finding aligns with a prior study conducted in Bangladesh [5]. Similar observations were reported by Humayoun et al [26]. in Pakistan, R. P. Khetan et al [34]. in Nepal, and R. Unnikrishnan et al [35]. in India. However, Kuna et al [36]. in Poland and A. Nigam [37] in India found thrombocytopenia in 20% and 60% of patients, respectively, indicating a comparatively lower prevalence than what was observed in our study. Although the mechanism of thrombocytopenia in dengue involves bone marrow destruction and peripheral platelet degradation, the precise cause remains unclear.

Leukopenia was noted in 32.5% of the patients in our study, consistent with a prior study in Bangladesh, which reported a higher prevalence compared to Kuna et al.'s findings (24.6%) [36]. Elevated hematocrit levels and increased levels of AST and ALT (>45 IU/L) were also observed in our research, corroborating findings by Humayoun et al [26]. The clinical features and laboratory parameters, including hematological and biochemical findings, play a crucial role in the prompt management of dengue fever. This study revealed that fever, nausea, vomiting, headache, abdominal pain, myalgia, skin rash, and diarrhea are the most prevalent features. Key indicators such as a high leukocyte count, low platelet count, elevated AST and ALT values, and an increased hematocrit value serve as vital parameters for the rapid diagnosis of dengue-infected patients.

Conclusion

This study shows majority of patients were in the recovery phase, emphasizing the various stages of Dengue Virus Infection observed in the tertiary care setting. Clinical features exhibited a diverse range

of symptoms, with fever being predominant. Complications, such as bleeding, pleural effusion, and breathlessness, were notable, emphasizing the complexity of Dengue cases. Laboratory parameters further highlighted the impact of Dengue, with thrombocytopenia and raised hematocrit levels being common abnormalities. The detection of NS1 antigen and seropositivity to NS1+IgM were crucial for early diagnosis, whereas the Tourniquet Test provided additional diagnostic value. This comprehensive examination sheds light on the multifaceted nature of Dengue Virus Infection at a tertiary care hospital, emphasizing the importance of prompt clinical intervention, continuous monitoring, and effective public health strategies to manage the diverse presentations of Dengue and mitigate its impact on the affected population.

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