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Clinical and Hematological Profiling of Dengue Fever: Impact of Age, Gender, and Comorbidities on Disease Severity

Muhammad Noman[1], Syeda Ruhma Rizvi[1], Amna Talib[1], Hafiza Sania Younas[2], Maryam Saeed Khan[3], Urwah Aybk[1], Momina Khalid[1], Munir Hussain[4]

[1]Medical Laboratory Technology (MLS), Department of Pathology, Faculty of Medicine and Allied Health Sciences, The University of Faisalabad (TUF), Faisalabad, Punjab, Pakistan.
[2]Institute of Microbiology, Faculty of Life Sciences, Government College University Faisalabad, Punjab, Pakistan.
[3]Department of Nutritional Sciences, Faculty of Medical Sciences, Government College University Faisalabad, Punjab, Pakistan.
[4]Medical Lab Sciences (MLS), Department of Pathology, Allied Hospital Faisalabad, Punjab, Pakistan.

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Correspondence to: Muhammad Noman Medical Laboratory Technology (MLS), Department of Pathology, Faculty of Medicine and Allied Health Sciences, The University of Faisalabad (TUF), Faisalabad, Punjab, Pakistan.
Email: Muhammadnoman.mls1@gmail.com

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ABSTRACT

This study analyzed 280 dengue patients (77.8% male, 22.1% female) with a median age of 30 years. The highest incidence occurred in the 40–60 age group (53%). Dengue fever (DF) was predominant (229 cases), while dengue hemorrhagic fever (DHF) affected 51 patients, with males showing higher infection rates (p=0.03). DHF patients were younger (median age 24.5 vs. 30 years) and exhibited severe symptoms like abdominal pain, rash, and elevated liver enzymes (AST/ALT levels 10x and 2.5x higher, respectively; p<0.05). Thrombocytopenia (<69,000 cells/mm^3) was significant in DHF (p=0.001). Common symptoms included myalgia (91%), headache (80%), and vomiting (52.5%). Hemorrhagic manifestations (e.g., epistaxis, melena) occurred in 50% of patients. Comorbidities (e.g., hepatitis C, diabetes) worsened outcomes, increasing DHF risk (32% vs. 20%; p=0.01) and mortality (CFR=2.5%). Serological testing confirmed positivity for NS1 (41.7%), IgG (30%), and IgM (27.8%).

INTRODUCTION

Dengue fever has remained a significant public health threat worldwide for the past three decades. Each year, nearly one billion infections are recorded worldwide, with as many as 2% leading to deaths. Such outbreaks continue to put regional healthcare systems and economies under stress, especially in developing nations, where they have become increasingly common. Dengue fever is a viral infection transmitted to humans by Aedes aegypti mosquitoes. Asia is one of the most vulnerable areas for its spread. Its global stress is expected to be comparable to that of tuberculosis and malaria in the future, posing substantial financial difficulties for populations and governing bodies (Nandwani, Bhakhri, Singh, Rai, & Singh, 2021).

Since 1992, Pakistan has seen an increasing number of dengue-related outbreaks, and the disease has grown into a seasonal nightmare in recent years. Although understanding the geographic distribution of diseases is critical for the development, execution, and evaluation of control programme, it is also crucial to understand the factors responsible for differences in the clinical signs among different groups of people (Freifeld, Mandl, Reis, & Brownstein, 2008). Research has consistently demonstrated that susceptibility to classic dengue fever and dengue hemorrhagic fever varies according to factors such as age, gender, and other socio-demographic characteristics within a given population. Understanding the relationship between the host and the disease is essential in developing strategies to control dengue fever (A. Ali et al., 2013). The purpose of this study was to identify the risk factors for dengue fever among patients seeking treatment at a tertiary care hospital. The results of this study can inform the development of targeted

preventive and control measures for the disease in the future.

Dengue affects individuals from diverse sociodemographic backgrounds and causes significant illness and mortality in children (Ho, Wang, Lin, & Liu, 2013). Children with dengue can experience a range of clinical outcomes, from mild illness to potentially fatal hemorrhage and shock. Researchers worldwide are attempting to identify the factors that determine the expected clinical course of the disease in order to make the most cost-effective use of available resources. The potential determinants include various clinical characteristics (Pandey et al., 2015), laboratory parameters (Wakimoto, Camacho, Guaraldo, Damasceno, & Brasil, 2015), biomarkers, and gene expression, each with varying degrees of clinical relevance. Thrombocytopenia and Hemocrit/hemoglobin Concentration markers have emerged as extensively researched and widely employed hematological parameters in the realm of clinical decision-making (Jayadas, Kumanan, Arasaratnam, Gajapathy, & Surendran, 2019).

Every year, it is estimated that approximately 390 million people come in contact with one of the four different types of the virus that causes dengue, i.e., (DENV1- DENV4), transmitted by mosquitoes. Viral infections can manifest as either asymptomatic cases or give rise to dengue fever or dengue alongside signs of infection, both of which are self-limiting infectious diseases (Organization et al., 2009). However, the disease progresses to severe dengue fever or DHF (dengue hemorrhagic fever) and possibly DSS (dengue shock syndrome) in approximately 20-25% of symptomatic cases (Bhatt et al., 2013), resulting in probably fatal signs and symptoms (Organization et al., 2009). Severe dengue is characterized by endothelial dysfunction, which leads to blood vessel permeability along with leakage of plasma. This is thought to be due to an increase in the amount of circulating inflammatory agents like TNF-a and IL-1 (Malavige & Ogg, 2017). Despite ongoing attempts, there are actually no currently available techniques for predicting or preventing disease progression. As a result, understanding the mechanism(s) which control the excessive inflammation provoked by DENV is critical for effective management of serious cases and understanding its pathogenesis.

The primary objective of this study is to investigate the clinical and epidemiological landscape of dengue fever in Faisalabad, Pakistan, a region recurrently affected by dengue outbreaks. Specifically, the research seeks to analyze the demographic and clinical profiles of hospitalized dengue patients between January 2021 and January 2022, and to assess the role of comorbidities in exacerbating disease severity. Additionally, this study aims to identify the circulating dengue virus serotypes within this timeframe using molecular diagnostics, such as polymerase chain reaction (PCR), to elucidate the virological diversity contributing to disease transmission and progression. A critical focus of the research is to evaluate the diagnostic and prognostic utility of bedside markers—such as hematological and biochemical indicators—to enhance early detection and clinical management of dengue infections. By integrating virological, clinical, and environmental data, the study intends to contribute to the development of predictive models that support timely public health interventions and targeted strategies to mitigate the impact of dengue fever in endemic urban settings.

MATERIALS AND METHODS

Sample Collection and Study Setting

This experimental study was carried out over six months at the Isolation and Infectious Diseases Ward of Allied Hospital, Faisalabad. A total of 280 blood samples were collected from patients presenting with clinical suspicion of dengue fever. The inclusion criteria specified patients with a history of fever lasting between two and ten days, accompanied by relevant clinical symptoms and hematological markers, such as thrombocytopenia and leukocytopenia. All age groups and genders were included, provided they had a supporting travel history or exposure to endemic areas. The classical venipuncture technique was used to collect 3–5 ml of venous blood, which was processed immediately for diagnostic evaluation. Patients were excluded if they presented with fever outside the 3–10 day window, exhibited normal CBC results, or had a known history of chronic illnesses, in alignment with WHO diagnostic recommendations (World Health Organization [WHO], 2009).

Patient Information and Questionnaire

A structured questionnaire (Performa) was administered to each patient visiting the medical emergency dengue counter. The Performa gathered detailed demographic, anthropometric, and clinical data, including age, gender, weight, BMI, medication history, prior dengue infection, comorbidities, travel history, and overall health condition. This systematic data collection facilitated comprehensive correlation between clinical symptoms and laboratory findings, enhancing diagnostic precision and allowing for the identification of risk factors and disease patterns (Kularatne, 2011).

Epidemiological Background

The study design was informed by the historical context of the 2011–2012 dengue outbreak in Pakistan, which had a significant impact in urban areas such as Lahore and Faisalabad. Hospital-based reports and existing literature were reviewed to assess the frequency and severity of dengue cases during this period. This background information guided both patient selection and the interpretation of diagnostic data about prevailing regional trends. Sporadic outbreaks occurring after the major epidemic were also considered for comparative evaluation of epidemiological shifts (Rashid et al., 2013).

Diagnostic Criteria for Dengue Classification

Dengue diagnosis was based on the revised WHO classification system, which categorizes cases as Dengue Fever (DF) or Dengue Hemorrhagic Fever (DHF). DF was defined by the presence of a 2–7 day fever and at least two symptoms such as retro-orbital pain, rash, headache, or myalgia. DHF was characterized by bleeding tendencies, evidence of plasma leakage, and marked

thrombocytopenia (platelet count <100,000/mm^3). Confirmation of dengue infection requires laboratory detection of NS1 antigen or IgM/IgG antibodies, particularly after the fifth day of illness (WHO, 2009; Simmons et al., 2012).

Clinical Examination and Data Collection

Upon the patient's initial hospital visit, a thorough clinical examination was conducted to assess general health, BMI, and signs of systemic illness. Detailed inquiries were made regarding travel history, contact with known cases, previous infections, and the presence of chronic diseases. Following clinical evaluation, blood samples were collected for hematological, biochemical, and immunological analyses. Additional diagnostic support was provided through radiological and coagulation assessments. This comprehensive evaluation facilitated a stepwise confirmation of dengue cases, differentiating between suspected and confirmed infections (Bhatt et al., 2013).

Hematological and Biochemical Investigations

Hematological analysis was performed using an automated hematology analyzer (Swelab Alpha Plus) on EDTA-anticoagulated blood. Key parameters assessed included RBC count, WBC count, hemoglobin concentration, hematocrit, platelet count, and differential leukocyte count (neutrophils, lymphocytes, eosinophils, monocytes, and basophils). Abnormalities, such as thrombocytopenia, leukopenia, or erythrocytopenia, were flagged according to established reference ranges. For biochemical analysis, serum levels of liver enzymes alanine transaminase (ALT) and aspartate transaminase (AST) were measured using the kinetic method on a semi-automated analyzer (Merck Microlab 300), adhering to the protocols defined by the International Federation of Clinical Chemistry (IFCC) (Kuo et al., 1992; Wang et al., 2016).

Laboratory Diagnosis and ELISA Testing

To confirm dengue infection, blood samples were tested using commercially available ELISA kits for NS1 antigen and dengue-specific IgM and IgG antibodies. The presence and concentration of these immunological markers were used to estimate the stage of infection and disease severity. Molecular techniques, such as real-time polymerase chain reaction (RT-PCR), were employed selectively for dengue virus serotyping and viral load quantification, providing additional insight into the circulating dengue virus serotypes and their pathogenic potential (Gubler, 1998; Guzman & Harris, 2015).

Blood Sample Handling and Storage Protocols

Blood was collected using sterile Luer Lock syringes and distributed into EDTA and gel vacutainer tubes. Strict aseptic protocols were followed to minimize the risk of contamination. Samples were transported to the laboratory within 24 hours and maintained at a temperature between 2°C and 8°C. Serum separation was performed via centrifugation. For delayed testing, serum and plasma samples were stored at either -20°C or -70°C, with efforts made to minimize multiple freeze-thaw cycles, thereby preserving protein and antibody integrity (Rodriguez-Roche & Gould, 2013).

Radiological Examination for Diagnostic Support

Radiological assessments were conducted when clinically indicated, primarily to detect pleural effusion, which is a common complication in severe dengue cases. Chest X-rays were obtained for patients with respiratory symptoms or clinical suspicion of fluid accumulation. Although not a standard diagnostic tool for dengue, radiological evidence was used to support differential diagnosis and assess disease severity, particularly in cases that progressed toward Dengue Hemorrhagic Fever (Parkash et al., 2015).

RESULTS

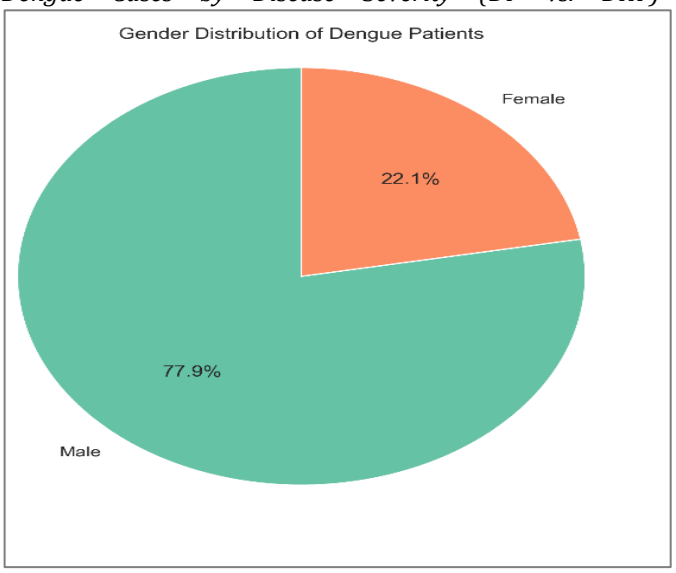
Clinical Characteristics of patients

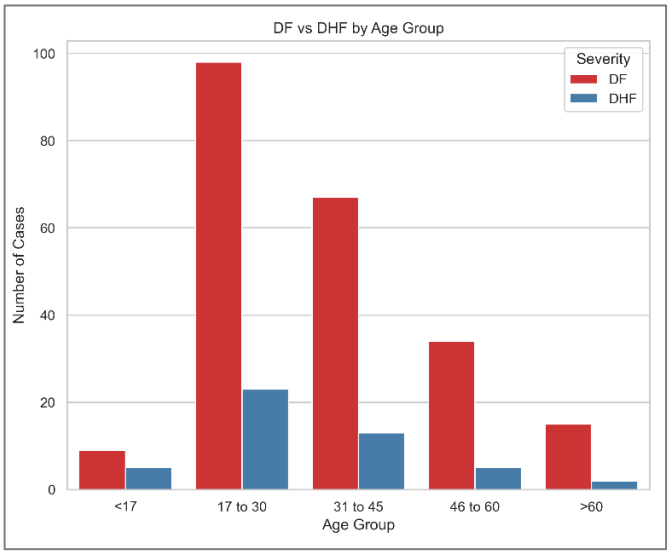
Among 280 individuals under observation, 218 (77.8%) were identified as male, while the remaining 62 (22.1%) were identified as female, as illustrated in **Figure 1(A).** The median age recorded among these patients was 30 years, with age variations spanning from 6 to 90 years. In the studied population, it was observed that three individuals (1%) fell within the age range of 6 to 16 years, while the majority consisted of 277 adults (98%). Notably, it was observed that no children under the age of 6 were enrolled during the two-year period under examination. Among the adult patients, there was a total of 90 individuals (32%) aged between 17 and 30 years. The age group with the highest occurrence of dengue patients was between 40 and 60 years, accounting for 53% (153) of the cases. Additionally, the age group spanning 17 to 39 years represented 45% of the affected individuals. Examining the data over the years, it becomes evident that the demographics of individuals affected by dengue have shown a rise in the number of elderly patients in 2021, compared to preceding years. Significantly, there was a noticeable surge in the median age of individuals, reaching 21.5% years in 2021. Moreover, there was a decline in the incidence rate of dengue patients, specifically by 5%, among the 6- to 15-year-old age bracket in the same year. This data shows the distribution of dengue patients by age from January 2021 to January 2022.

DF was identified in a total of 229 individuals, comprising 179 males and 50 females. Similarly, DHF was diagnosed in 51 patients, with 39 males and 12 females affected (illustrated in Figure 4.3). It is noteworthy that a statistically significant difference (p-value = 0.03) was observed in the prevalence of DENV infection between males and females, with a higher occurrence among the male population. According to the data analysis, a notable difference was observed in the age distribution between patients diagnosed with severe dengue (DHF) and those with dengue fever (DF) (p-value = 0.05). It was observed that DF patients had a median age of 30 years, whereas DHF patients had a median age of 24.5 years. In the age bracket of 6-17 years, there witnessed a notable rise of 20% in the occurrence of individuals affected by dengue, alongside a significant increase of 23.9% in cases of dengue hemorrhagic fever (DHF) within the age range of 6 to 30 years, comparing the data from previous years data. Detailed breakdown of dengue cases based on disease severity and age groups is listed in **Figure 1(B).**

Figure 1

*Demographic and Clinical Distribution of Dengue Patients (n = 280): **1A.** Gender Distribution Among Dengue Patients: A Predominantly Male Cohort: **1B. Age** Stratification of Dengue Cases by Disease Severity (DF vs. DHF)*





Clinical Signs and Symptoms

In this study, various symptoms were observed in our study participants. Myalgia was reported in 255 individuals, accounting for 91% of the participants. Headache was experienced by 225 patients, representing 80% of the sample, while vomiting occurred in 147 individuals (52.5%). Other symptoms included abdominal pain in 40 patients (14.2%), nausea in 49 individuals (17.5%), cough in 25 patients (8.9%), rash in 22 participants (7.85%), retro-orbital pain in 22 cases (7.85%), sore throat in 18 individuals (6.42%), anorexia in 15 patients (5.3%), restlessness in 14 individuals (5%), diarrhea in 12 patients (4.2%), periorbital puffiness in 8 cases (2.85%), and cold/clammy skin in 6 individuals (2.14%) as detail mentioned in **Table 1** and **Figure 2(A)** for DF patients and **Figure 2(B)** for DHF patients for further information).

Table 1

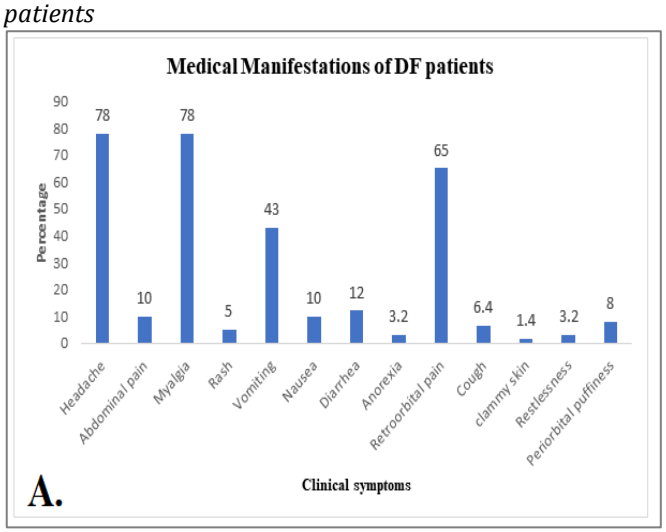
The medical manifestations of DF and DHF patients

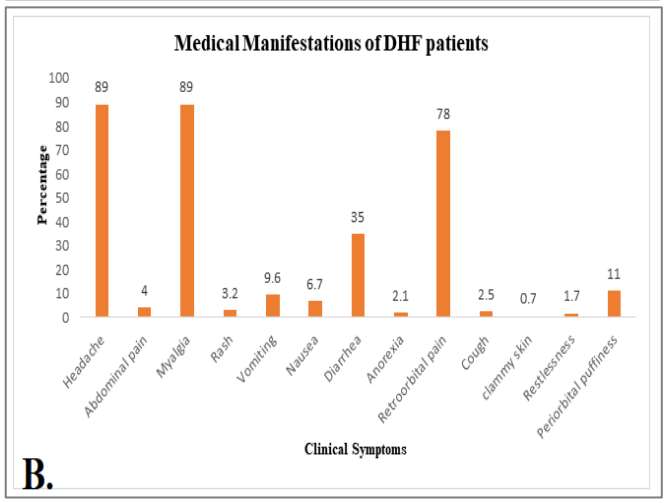
Characteristics	DF (%)	DHF (%)
Headache	185 (66)	40 (14)
Abdominal pain	29 (10)	11 (4)
Myalgia	175 (62)	50 (18)
Rash	13 (5)	9 (3.2)
Vomiting	120 (43)	27 (9.6)
Nausea	30 (10)	19 (6.7)
Diarrhea	8 (2.8)	4 (1.5)
Anorexia	9 (3.2)	6 (2.1)
Frontal headache	14 (5)	8 (2.8)
Cough	18 (6.4)	7 (2.5)
clammy skin	4 (1.4)	2 (0.7)
Restlessness	9 (3.2)	5 (1.7)
Periorbital puffiness	5 (1.7)	3 (1)

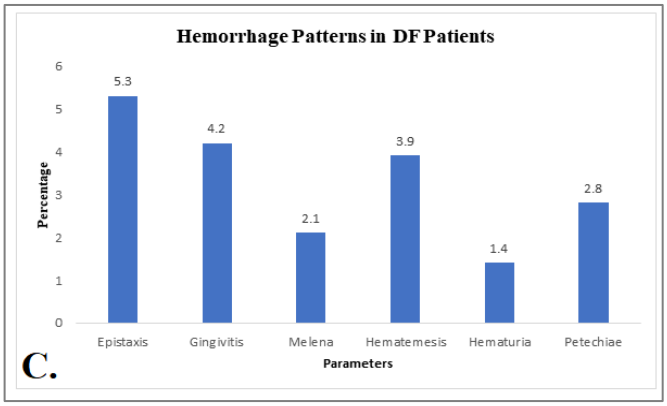
Among the total of 280 individuals assessed, approximately 50% or 140 patients experienced diverse forms of hemorrhage. The specific types observed comprised epistaxis, affecting 24 patients (8.5%), followed by gingivitis in 22 individuals (7.8%), melena in 16 cases (5.7%), hematemesis in 17 occurrences (6%), petechiae in 13 instances (4.6%), and hematuria in 6 patients (2.1%). The incidence of spleen enlargement was found in 29 individuals (9.7%), while liver enlargement was observed in 30 patients (10%). Additionally, abdominal tenderness was reported by 12 individuals (4.2%) as depicted in **Figures 2(C) and 2(D).**

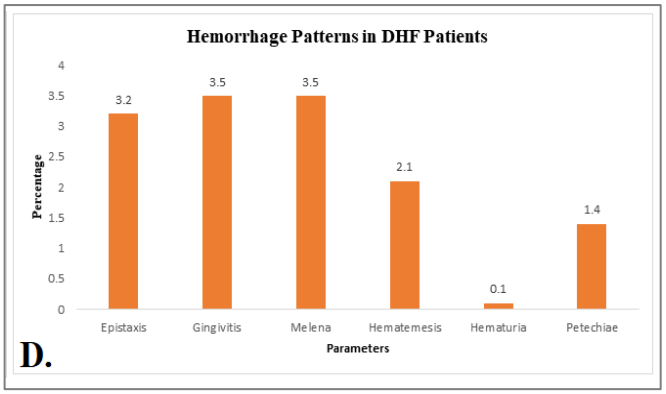
Figure 2

*Clinical Symptomatology and Hemorrhagic Manifestations in Dengue Fever and Dengue Hemorrhagic Fever Patients. **2(A):** Clinical Presentations of DF Patients. **2(B):** Clinical Presentations of DHF Patients. **2(C):** Comparative Evaluation of Hemorrhage Patterns in DF patients. **2(D):** Comparative Evaluation of Hemorrhage Patterns in DHF patients*









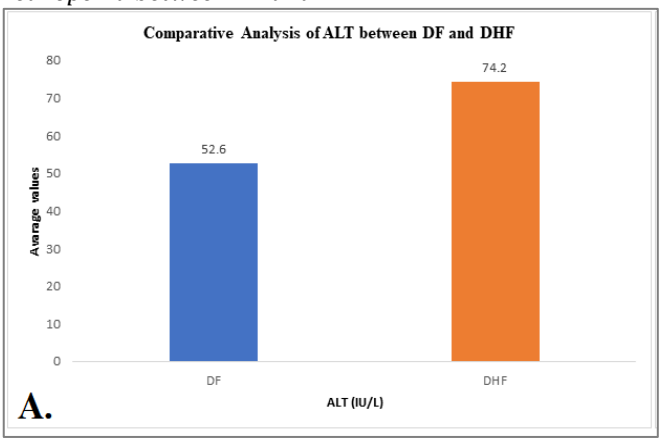
The most prevalent symptoms, including headache, myalgia/arthralgia, and vomiting, were observed among both DF and DHF patients, but no substantial disparities were detected between the two cohorts. Conversely, individuals afflicted with DHF did not exhibit any notable variations in comparison. Dengue hemorrhagic fever experienced distinct symptoms without any signs of plagiarism. Abdominal pain, rash, periorbital puffiness, nausea, and clammy skin exhibited a significant correlation with DHF cases, as indicated by a p-value of less than 0.05. While restlessness and frontal headache were more commonly observed in DHF cases, the association was not statistically significant. Moreover, spontaneous hemorrhages, abdominal tenderness, splenomegaly, and hepatomegaly were frequently observed in individuals with DHF, with a p-value of less than 0.05.

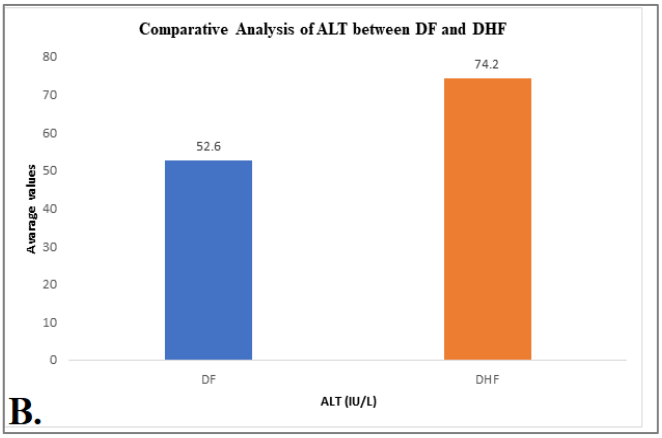
Laboratory Findings

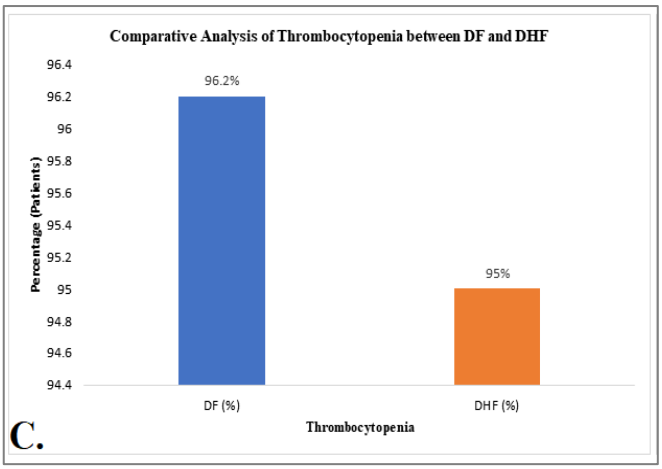
Liver involvement, as evidenced by atypical liver enzyme levels, was detected in different individuals diagnosed with dengue fever, as detailed in **Figure 3(A-D)**. Specifically, alanine aminotransferase (ALT) levels were elevated in 53% of cases, while aspartate aminotransferase (AST) levels were elevated in 65% of the patients. The occurrence of elevated liver enzyme levels, specifically aspartate aminotransferase (AST) and alanine aminotransferase (ALT), was predominantly observed in cases of dengue hemorrhagic fever (DHF). In 70% of DHF patients, ALT levels were found to be elevated, as indicated by a statistically significant p-value of 0.032. Similarly, all individuals with DHF exhibited increased AST levels with a statistically significant p-value of 0.037. The levels of ALT were found to be 2.5 times greater than the standard values in patients diagnosed with DHF, whereas AST levels were 10 times higher. Upon admission, a significant majority of the DHF patients (95%) exhibited thrombocytopenia. Increased hemoglobin was noted in 64% and 76% of individuals with DF and DHF, respectively, and reduced white blood cell count was seen in 72% and 70% of individuals with DF and DHF, respectively. The presence of raised Hb is frequently linked (p-value<0.05) to DHF, which serves as a reliable marker. Remarkably, during hospitalization, DHF patients (81%) exhibited a significantly higher hematocrit level compared to DF patients (67%).

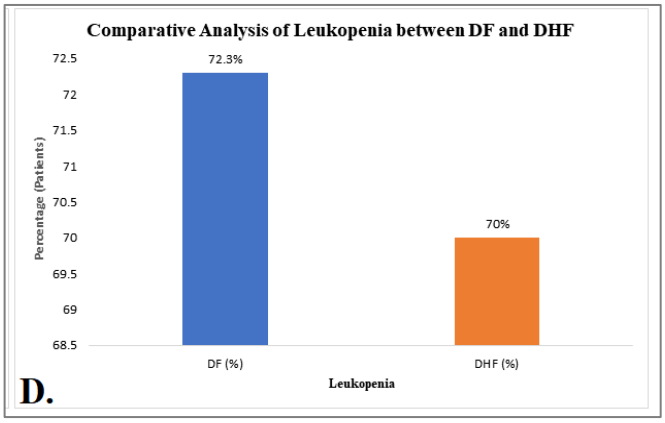
Figure 3

Comparative Analysis of Biochemical and Hematological Parameters in Dengue Fever and Dengue Hemorrhagic Fever Patients. ***3(A):*** *Comparative study of ALT between DF and DHF.* ***3(B):*** *Comparative study of AST between DF and DHF.* ***3(C):*** *Comparative study of Thrombocytopenia between DF and DHF.* ***3(D):*** *Comparative study of Leukopenia between DF and DHF*









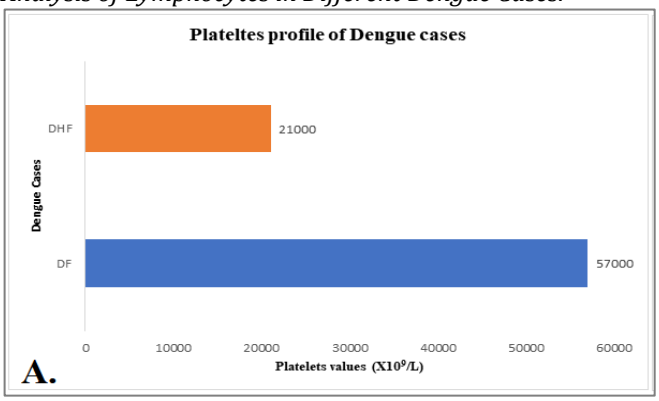
Hematological Parameters (CBC)

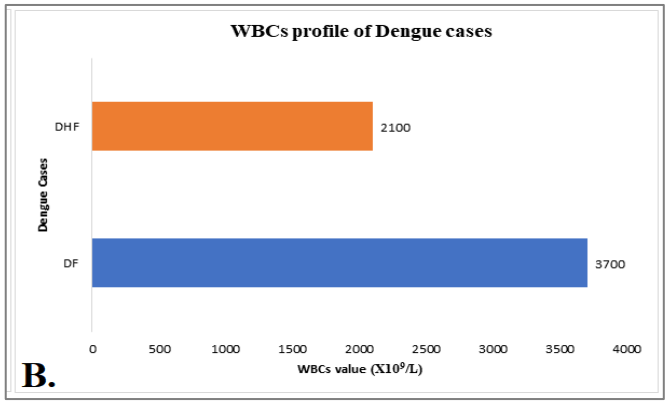
In our research, a comprehensive analysis was conducted on a sample size consisting of 280 participants, who were classified into two main groups: Dengue Fever (DF) and Dengue Hemorrhagic Fever (DHF). It is essential to note that both DF and DHF have the potential to induce significant alterations in various blood parameters. An analysis was conducted on the peripheral blood parameters recorded from blood specimens of patients, specifically focusing on the data collected during the first to third day (acute and febrile phase) and the fifth day (critical phase). This investigation examined the variations in these parameters between individuals who experienced leakage and those who did not, specifically during the initial stage of the infection (days 2 and 3) as detailed in **Figure 4(A to G)**.

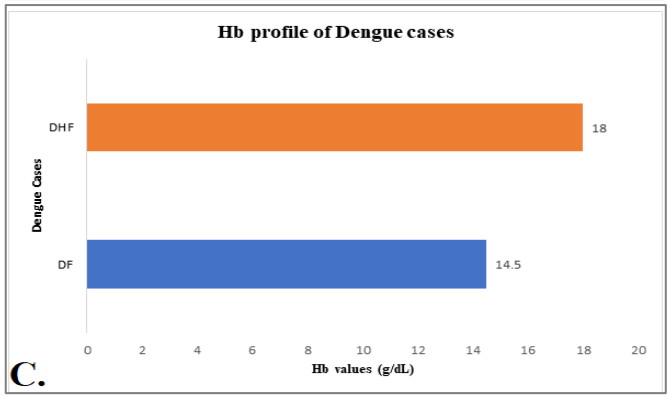
Leucopenia was found to have a p-value of 0.198 for patients with DF and DHF. Remarkably, a notable correlation was detected between Hemoglobin and HCT, with p-values of 0.046 and 0.369, respectively. Thrombocytopenia, defined as a platelet count below 69,000 cells/mm3, was found to have a p-value of 0.001, as depicted in **Table 2**. The correlation between neutrophils and lymphocytes and the stage of the disease was not statistically significant.

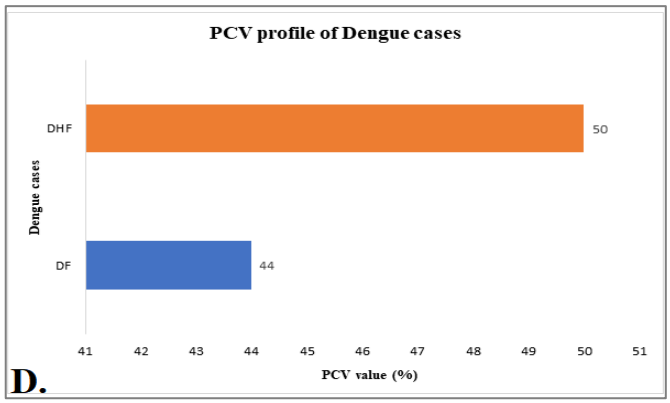
Figure 4

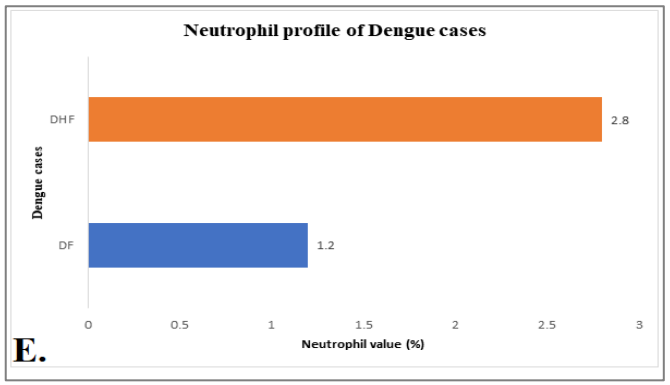
Comparative Hematological Profiling in Diverse Clinical Presentations of Dengue Infection. ***(A)*** *Comparative Analysis of Platelet Values in Different Dengue Cases.* ***4(B):*** *Comparative Analysis of WBC Values in Different Dengue Cases.* ***4(C):*** *Comparative Analysis of Hb Profile in Different Dengue Cases.* ***4(D):*** *Comparative Analysis of PCV Values in Different Dengue Cases.* ***4(F):*** *Comparative Analysis of Neutrophils in Different Dengue Cases.* ***4(G):*** *Comparative Analysis of Lymphocytes in Different Dengue Cases.*











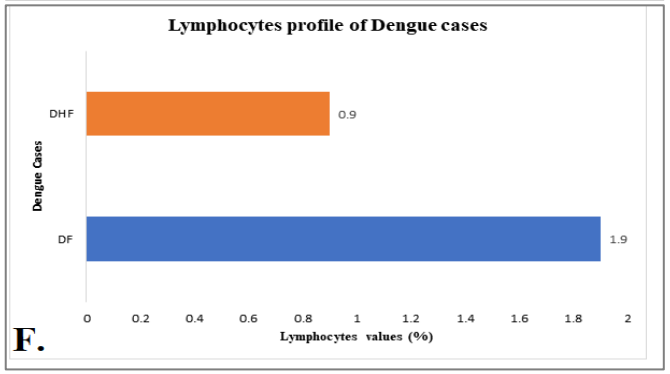


Table 2

Hematological Profile of a Patient Affected by Dengue Fever

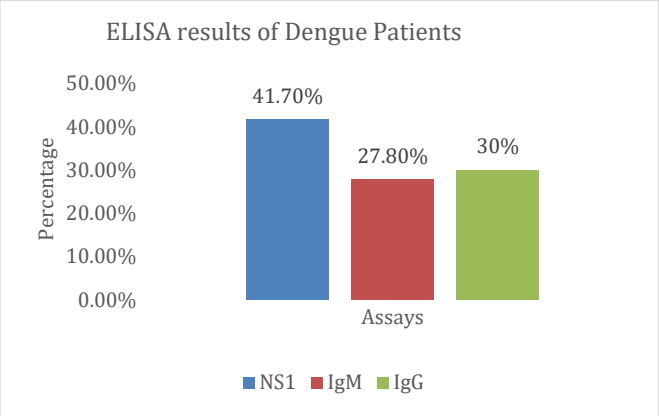
CBC Parameters	Reference Values	Dengue cases	
		DF	DHF
Platelets (x10^9/L)	300000	57000	21000
WBCs (x10^9/L)	8000	3700	2100
Hb (g/dL)	13	14.5	18
PCV (%)	39	44	50
Lymphocytes (%)	30	1.9	0.9
Neutrophils (%)	60	2.8	1.2

ELISA (NS1, IgM, IgG)

Through the use of diverse assays to detect the presence of the dengue virus, a comprehensive analysis was conducted on the blood samples of 280 individuals who exhibited symptoms suggestive of dengue fever.

Figure 5

A comparative analysis of IgG, IgM and NS1 in dengue patients



These samples were subjected to distinct detection methodologies, resulting in the confirmation of 280 cases as positive for dengue infection. The individuals under investigation underwent evaluations using NS1, IgM, and IgG methods, resulting in the confirmation of dengue-positive cases in 71.5% of the suspects. Among them, 117 subjects (41.7%) tested positive for NS1, while 85 individuals (30%) exhibited positive results for IgG. Additionally, 78 individuals were found to be positive for anti-dengue IgM antibodies as detailed in **Figure 5**.

Comorbidities in Patients with Dengue Fever

Several intrinsic (host/physiological) and extrinsic (viral) factors influence the course of DENV infection. These factors encompass different elements, including the individual's prior exposure or dengue immune status during the current infection with dengue (Dinh The Trung & Wills, 2014). Furthermore, the specific dengue virus serotype 2, advanced age (over 65 years) (M.-S. Lee, Hwang, Chen, Lu, & Chen, 2006), the Asian genotype of the DENV, certain ethnic backgrounds (Boillat-Blanco et al., 2018)Infection in children and infants who contract transplacental infection with DENV throughout pregnancy is also a factor. Furthermore, the presence of concurrent health conditions in individuals with dengue fever is acknowledged as a significant determinant for the onset of severe disease manifestations, such as DHF (dengue hemorrhagic fever) and DSS (dengue shock syndrome), as well as for the prolongation of hospitalization periods. This correlation significantly contributes to the escalation of morbidity, mortality rates, and the overall burden on public health systems within countries where dengue fever is prevalent (Willeam Peter et al., 2019).

The case fatality rate (CFR) is a metric that gauges the gravity of diseases, measuring the proportion of fatalities within a specified population afflicted by a particular disease or condition during a designated timeframe. The case fatality rate (CFR) experiences a significant rise when individuals have comorbidities, and this effect becomes even more pronounced when combined with severe dengue (Werneck et al., 2018). Hence, addressing the coexisting medical conditions poses a significant obstacle in minimizing the number of deaths associated with dengue fever. It is essential to highlight that comprehensive and consistent information is scarce regarding the impact of pre-existing comorbidities on the prognosis of dengue infection worldwide. In 2021, a thorough investigation was conducted in Faisalabad to assess the impact of pre-existing medical conditions on the progression and severity of dengue among patients admitted to the hospital. This study focused on individuals affected by dengue and concurrent comorbidities, aiming to analyze the potential exacerbation of the disease caused by these underlying health issues.

Patients' characteristics

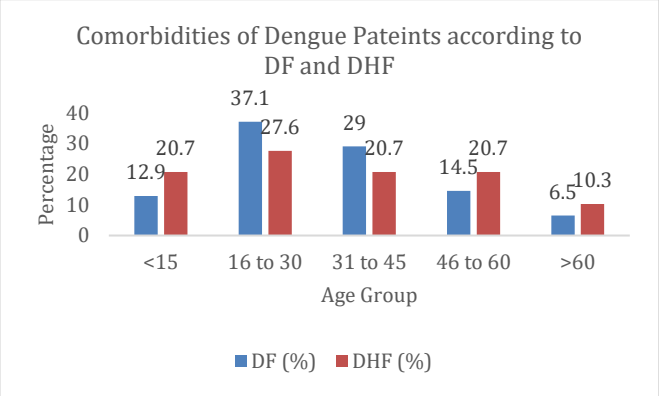
A total of 82 individuals out of 280 suspected patients diagnosed with dengue, presenting either single or multiple comorbidities, were included in the study. However, the data of 5 patients (5.1%) had to be excluded due to incomplete information, as they decided to leave the hospital against medical advice (LAMA). A total of 93 individuals diagnosed with dengue fever participated in the study, comprising 65 (69.8%) males and 28 (28.5%) females. The patients' age ranged from 4 to 90 years, with a median age of 31 years. Among them, 10 (10.2%) were children, aged 16 years or below, while 88 (89.7%) were adults. The majority of individuals seeking medical care (40%) fell within the 40- to 60-year age bracket, while 22% were classified within the 17- to 30-year age group. The smallest proportion of patients (7%) were under the age of 10 years. The incidence of diseases exhibited a steady rise in correlation with age, reaching its peak within the age groups of 46 years and older. This trend was mirrored in the distribution of gender across these particular age brackets.

In **Figure 6,** the gender distribution of individuals affected by dengue is depicted across various age groups. Out of the total number of patients, 62 (68.1%) were diagnosed with DF, including 50 (80.7%) males and 12 (19.3%) females. Additionally, 29 (31.9%) patients were diagnosed with DHF, comprising 19 males (65.5%) and 10 females (34.5%). The DF patient group had a median age of 32, whereas the DHF patient group had a median age of 31. Nevertheless, no statistically significant difference was observed between the ages of DF and DHF patients (p-value = 0.9). The categorization of dengue patients into various age groups based on disease severity did not reveal any notable distinctions among them (p-value = 0.61). The age distribution of dengue patients with accompanying medical conditions. Likewise, no significant disparity (p-value = 0.12) was observed in the gender distribution between individuals diagnosed with Dengue

Fever (DF) and those with Dengue Hemorrhagic Fever (DHF).

Figure 6

Categorization of Dengue Patients with Co-morbidities by Age Group



Prevalence of Comorbidities among Dengue Patients

Numerous patients who were documented with DENV infection presented with a range of individual or concurrent comorbidities. Out of the 98 cases examined, a total of 75 (76.5%) patients were found to have a single comorbidity, while 15 (15.3%) individuals exhibited two comorbidities. Furthermore, a smaller subset of 8 (8.16%) patients were diagnosed with three distinct comorbidities. Furthermore, a solitary instance (1.1%) was identified with six underlying medical conditions. Among the prevalent comorbidities reported in dengue patients, numerous infectious ailments such as typhoid, pneumonia, viral hepatitis, TB, malaria, septicemia, and UTI took precedence. Additionally, non-communicable diseases such as cancer, hypertension, diabetes mellitus, and ischemic heart disease were also commonly observed in individuals diagnosed with dengue.

In **Table 3**, an overview is provided of the prevalence of different comorbidities and medical conditions observed in individuals diagnosed with dengue. Co-occurring infections revealed that the hepatitis C virus was the most frequently documented, affecting 45 patients, accounting for 45.9% of the cases. Additionally, the malarial parasite (MP) was found in 6 patients (6.1%), while typhoid was identified in 10 patients (10.2%). In every instance, cases of hepatitis A, hepatitis B, and urinary tract infection were documented. In terms of non-communicable diseases, diabetes mellitus was observed in 5 (5.1%) patients, while hypertension affected 3 (3%) patients.

Table 3

Frequency of different co-morbidities in dengue patients

Comorbidities and Medical Manifestation	Frequency N= 82 (%)
Hepatitis C	45 (45.9)
Typhoid	10 (10.2)
Malaria	6 (6.1)
Diabetes Mellitus (DM)	5 (5.1)
Hypertension	3 (3)
Urinary Tract Infection (UTI)	1 (1.1)
Hepatitis B, Hepatitis C	1 (1.1)
Typhoid, Hepatitis C	1 (1.1)
Malaria, Typhoid	1 (1.1)
Hypertension, Diabetes	4 (4.4)
Ischemic Heart Disease, Diabetes	2 (2.2)
Hepatitis B, Malaria, Hepatitis C	1 (1.1)
Hepatitis C, Hypertension, Diabetes	1 (1.1)
UTI, Hypertension	1 (1.1)

Moreover, apart from these isolated instances of diverse comorbidities, there were several instances of multiple co-infections and/or non-communicable diseases (NCDs). Notably, individual cases of hepatitis C co-occurring with hepatitis B, hepatitis C along with typhoid, hepatitis C and malaria, hepatitis B and malaria, typhoid and malaria, as well as septicemia combined with aspiration pneumonia were documented. The occurrence of comorbidities was observed in the patient population under study. Specifically, a combination of diabetes and ischemic heart disease was noted in two individuals, accounting for 2% of the cases. Another case involved the coexistence of diabetes and hypertension, reported in one patient, representing 1.1% of the sample. Similarly, hypertension combined with ischemic heart disease was observed in one patient, also accounting for 1.1.

The outcome of DF with Comorbidities

A thorough investigation was conducted to compare the information regarding dengue patients who had comorbidities, focusing on the variation in signs, symptoms, and treatment outcomes based on the severity of the disease (DF vs. DHF). Moreover, a comparative analysis was conducted to investigate the likelihood of developing severe dengue in patients with and without comorbidities based on the reported data. The examination revealed that the presence of additional medical conditions during a dengue infection significantly contributed to the worsening of the illness. The incidence of severe dengue fever (DHF/DSS) was significantly higher (p-value=0.01) in individuals diagnosed with dengue and comorbidities (32%) compared to those without any underlying health conditions (20%). Likewise, patients with underlying medical conditions exhibited a notable occurrence of significant internal bleeding, as indicated by melena, hematemesis, hematuria, and various other manifestations related to hemorrhaging. Nevertheless, there was no evident correlation observed between the indications and symptoms of the ailment within either of the groups. Remarkably, individuals suffering from dengue, who did not have any underlying health conditions, experienced a higher prevalence of distinct clinical manifestations such as headaches, myalgia, and rashes, as indicated by statistically significant findings (p-value<0.05). Conversely, dengue patients with comorbidities exhibited a comparatively lower incidence of these symptoms. The occurrence of diarrhea, anorexia, cough, and periorbital puffiness showed a higher prevalence, although not statistically significant, among patients with comorbidities. In contrast, patients with comorbidities exhibited a greater frequency of splenomegaly, hepatomegaly, and severe thrombocytopenia (platelet count <15,000 cells/mm3) when compared to those with DF alone, with a p-value less than 0.05.

The prevalence of low hemoglobin levels was more frequently observed in patients with co-existing medical conditions, although the difference was not statistically significant (p-value=0.09), compared to those with dengue

infection alone. An exclusive analysis was conducted to compare the data of patients who developed severe disease, specifically dengue hemorrhagic fever (DHF), with or without underlying comorbidities. Interestingly, similar patterns were noted in terms of the clinical symptoms and hemorrhagic manifestations of the disease. Additionally, the data of patients diagnosed with concurrent infections such as hepatitis C and malaria was also compared and will be elaborated upon below.

Dengue Fever and Hepatitis C

Among the multiple medical conditions, 53 (18%) had concurrent infections with HCV including 11 females and 42 males. Out of these 48 (17%) had solely HCV infection. there was one patient with ischemic heart disease, hypertension, diabetes mellitus, abdominal tuberculosis, and typhoid. Additionally, one patient had co-infection with hepatitis B virus (HBV), one with malaria, one with typhoid, one with HBV and another with co-occurrence of diabetes and hypertension. There was no statistically significant variation among gender, age, symptoms and signs, and medical outcome among individuals who had or did not have HCV concurrent infections. Individuals with HCV concurrent infections, on the other hand, had considerably greater hemorrhagic behaviors. Data from 48 individuals with dengue (38 males and 10 females) who were only associated with HCV were compared with those who had only a dengue infection. DHF was identified in 16 (33.3%) of the 32 (66.7%) HCV-associated dengue patients. Surprisingly, DENV-HCV co-infected individuals who developed DHF were significantly older (45 years) compared to those having dengue alone (30 years).

Case Fertility Rate (CFR)

There was a total of 12 fatalities documented, resulting in an overall case fatality rate of 2.5%, specifically attributed to the severe manifestation of dengue fever, known as DHF/DSS. The individuals affected had a range of pre-existing health conditions and predominantly succumbed to the consequences of multiple organ failure. The case fatality rate (CFR) was determined by considering the overall count of confirmed dengue cases (n = 380), encompassing individuals diagnosed with dengue infection, regardless of their refusal to take part in the study, premature departure against medical advice, or lack of adequate data.

Independent sample t-test

The results of the independent t-test are presented in the table, which shows the values of various laboratory parameters for dengue-reactive and non-reactive patients. The parameters measured include Hb (Hemoglobin), Platelets, IgG, IgM, MP (Malaria Parasite), WBC (White Blood Cell), and HCT (Hematocrit), as detailed in **Table 4**. Each parameter is reported for non-reactive and reactive patients, along with the mean and standard deviation (SD) values and the corresponding p-values.

Table 4

Independent t-test results depending upon NS1

Study Parameter	NS1-reactive (n=261) Mean±SD	NS1-non-reactive (n=19) Mean±SD	*P* value
Hb	18.73±5.223	14.33±4.875	0.046
Platelets	32.79±14.432	112.83±15.179	0.001
IgG	1.72±.449	0.83±.408	0.003
IgM	1.69±.462	0.17±.408	0.007
MP	1.01±.083	1.00±.000	0.839
WBC	3143.75±544.614	6433.33±242.212	0.004
HCT	41.68±4.001	40.17±4.750	0.369
ALT	52.23± 5.083	34.76± 3.02	0.032
AST	68.57± 4.75	38.98± 4.09	0.037

DISCUSSION

Dengue virus poses a significant global health challenge, endangering approximately half of the world's population. As a result, it has emerged as a critical concern within the realm of public health, particularly in nations with tropical climates (Gurung, Karki, Khadka, Gurung, & Dhakal, 2022). Faisalabad city holds a prominent position as a prime destination for individuals seeking quality healthcare services, surpassing the healthcare facilities available in neighboring districts. In the densely populated central regions of Faisalabad city, cases of dengue patients were reported. Through an analysis of data spanning one-year, specific areas were identified as emergence hotspots, indicating a higher likelihood of new dengue cases emerging or re-emerging in those locations. Moreover, the emergence of fresh instances of dengue fever in neighboring localities to these focal points underscores the transmission of dengue infection to contiguous territories. Furthermore, the data from both years also revealed the outward expansion of dengue infection from densely populated central zones towards adjoining sparsely populated districts within the city. The dynamics of the dengue fever epidemic are shaped by intricate interplays between the virus, vector, host, and are directly reliant on climatic and environmental factors (Descloux et al., 2012).

During the entire research period, it was evident that the highest occurrence of DENV infection occurred between August and October. The presence of seasonal fluctuations in humidity and temperature holds a significant influence on the survival of disease-carrying vectors, thereby directly contributing to the emergence of dengue epidemics. This study provides clear evidence of a higher ratio of male victims to females among the affected patients. These findings align with a separate study conducted to analyze the incidence of dengue across six Asian countries, which also reported similar observations (Anker & Arima, 2011). Numerous studies carried out across various Asian regions have consistently reported analogous findings (Yew et al., 2009). The observations above have revealed distinctive variances in dengue incidence based on gender, potentially influenced by factors such as exposure to the dengue vector or other unidentified variables. Remarkably, reports on DENV infection rates in North America have indicated an equitable distribution between females and males, or in some instances, a higher proportion of females (Günther, Ramírez-Palacio, Pérez-Ishiwara, & Salas-Benito, 2009).

Various factors, including social, cultural, and exposure-related aspects may have influenced the occurrence of gender specificity in dengue infection. There is a disparity in the available information concerning the age group of individuals impacted by dengue. Previous investigations carried out in Asia, utilizing surveillance

data, established a connection between the age of afflicted individuals and the seriousness of the ailment.

In the present investigation, a comprehensive analysis was conducted on a sample of 280 participants, wherein it was determined that all individuals had contracted the dengue virus. Upon scrutinizing the entire cohort, it was noted that a prevalence of 77.8% was evident among male subjects, exceeding the prevalence observed in female patients, which amounted to 22.1%. The distribution of gender prevalence observed in our study aligns with the findings of previous research as reported by Khan and colleagues (J. Khan & Khan, 2015). According to our findings, the majority of the recorded instances (53%) occurred within the age bracket of 40-60 years, while (32%) of the patients belonged to the 17-30 years range. The elevated occurrence among older individuals can be linked to their greater exposure to the surrounding environment, thereby heightening the likelihood of encountering the vector. Additionally, the act of traveling for business and various other reasons may contribute to an elevated susceptibility to infection. Conversely, children and older individuals, who constitute the majority of other age groups, typically possess significantly lower levels of exposure to their surrounding environment. Furthermore, a preceding study has documented that the middle-aged demographic represents the population group that is most adversely impacted (Seneviratne, Malavige, & De Silva, 2006).

Additionally, the individuals exhibited characteristic signs of dengue infection, including pyrexia (100%), internal hemorrhaging, and hepatomegaly, affecting 30% of the patients. Based on a prior investigation, liver injuries observed in cases of dengue illness may arise from either the direct impact of the virus or the interaction between the host's immune response and liver cells. This interaction has the potential to contribute to the occurrence of vascular leakage within the liver (Shahid et al., 2017). Patients with secondary dengue infection exhibited a reduction in thrombocyte count, as indicated by the total blood parameters. This decline in thrombocyte levels was specifically observed in patients who tested positive for IgG antibodies. A notable distinction was noted among various phases of the illness and fluctuations in blood parameters. Prior research has indicated that platelet analysis can aid in the identification of fever stages such as DF and DHF (Ramirez-Ronda & Garcia, 1994). In simpler terms, the rise was noted in individuals who had experienced leakage and those who hadn't, as determined during the evaluation conducted on the fifth day of the illness. According to a comparable investigation, the classification of DHF can be accomplished by analyzing the hemoglobin density (S. Ali et al., 2019).

The discovery of diagnostic indicators to anticipate the occurrence of DHF (Dengue Hemorrhagic Fever) during dengue infection plays a crucial role in effectively managing the disease. Accurate identification of DHF relies on clinical manifestations that align with the criteria established by the World Health Organization (WHO) (Carlos et al., 2005). In the present investigation, various clinical characteristics and hematological irregularities exhibited notable distinctions between DF and DHF. These disparities encompass abdominal discomfort, queasiness, skin rash, chilly and damp skin, swelling around the eyes, nosebleeds, gum inflammation, dark stool, vomiting blood, blood in urine, enlargement of the spleen, and enlargement of the liver. Consequently, the amalgamation of these unusual manifestations could serve as potential indicators for the prediction of DHF (Carlos et al., 2005).

The current study has identified a novel finding known as periorbital puffiness, which pertains to the accumulation of fluid around the eyes, resulting in swelling of the orbits. It is noteworthy that this phenomenon demonstrates a significant association with DHF (Dengue Hemorrhagic Fever). Nonetheless, further investigation is necessary to determine the clinical significance of this indicator in assessing the severity of the disease. The majority of individuals participating in this research exhibited leucopenia, thrombocytopenia, heightened liver enzyme levels, and abnormalities in their blood composition. In patients with dengue hemorrhagic fever (DHF), the alanine aminotransferase (ALT) levels were found to be up to 2.7 times higher than the standard range. Furthermore, the aspartate aminotransferase (AST) levels were elevated to 10 times the normal range in all DHF patients, signifying their substantial role in predicting the severity of the disease. Liver injury is a prevalent occurrence in cases of DENV infection, where the virus infects hepatocytes and Kupffer cells, thereby mediating its effects (Souza et al., 2007). In recent times, the assessment of dengue virus infection severity has increasingly involved the utilization of ALT and AST levels. Several investigations have highlighted the ability of AST and ALT levels to serve as indicators for predicting the seriousness of the disease (Gulati & Maheshwari, 2007). Incredibly, the hematocrit levels, which serve as an indicator of capillary leakage, were found to be higher in patients diagnosed with Dengue Fever (DF) compared to those with Dengue Hemorrhagic Fever (DHF). This observation suggests that the occurrence of overt or occult bleeding could potentially contribute to this phenomenon.

CONCLUSION

The study aimed to identify and analyze the risk factors associated with dengue fever among patients at Allied Hospital, Faisalabad, over one year. The research employed a cross-sectional design, combining questionnaire surveys, medical records review, and laboratory tests to gather data on dengue cases and associated risk factors. The analysis of the data revealed several significant findings. Firstly, the study provided insights into the demographic characteristics of dengue patients. It was observed that individuals in younger age groups, particularly young adults, were more susceptible to dengue infection. Secondly, the study examined the relationship between dengue and environmental factors. It was found that temperature and rainfall were significantly associated with dengue transmission. Higher temperatures and increased rainfall were positively correlated with higher dengue incidence, indicating the importance of climate factors in the spread of the disease. These findings underscore the importance of implementing effective vector control measures, including eliminating breeding sites and promoting proper water

management, to reduce dengue transmission. Furthermore, it was found that public awareness campaigns and vector control activities, including insecticide spraying and larval source reduction, were associated with a decrease in the incidence of dengue cases. This highlights the effectiveness of proactive measures in controlling the disease and underscores the importance of continued investment in prevention strategies. The conclusions drawn from the study have significant implications for public health interventions and policy development. The findings emphasize the importance of targeted interventions focusing on vulnerable age groups, particularly children and young adults, to mitigate the impact of dengue fever. Moreover, the study relied on self-reported data and medical records, which may be subject to recall bias or incomplete information. In conclusion, the chapter provides a comprehensive overview of the study's findings on dengue-related risk factors among patients in Allied Hospital Faisalabad. The research contributes to the understanding of dengue transmission dynamics in the local context. It highlights the importance of demographic characteristics, the role of bedside markers, and preventive measures in controlling the disease. The findings have significant implications for public health interventions, underscoring the need for targeted strategies to reduce dengue incidence and enhance community health outcomes. Further research in this area is warranted to strengthen the evidence base and inform more effective dengue prevention and control efforts.

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