“STUDY OF PARTIAL THROMBOPLASTIN TIME AND PROTHROMBIN TIME AS A PREDICTOR OF BLEEDING IN DENGUE INFECTION “– ONE YEAR CROSS SECTIONAL STUDY

BY

REGISTRATION NO: BG0117006

Dissertation

Submitted to

KAHER, Belagavi, Karnataka

In partial fulfilment

of the requirements for the degree of

M .D.

IN

GENERAL MEDICINE

DEPARTMENT OF GENERAL MEDICINE

J. N. MEDICAL COLLEGE

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Endorsement by the HOD/ Principal/ Head of the Institution

This is to certify that the dissertation entitled **"STUDY OF PARTIAL THROMBOPLASTIN TIME AND PROTHROMBIN TIME AS A PREDICTOR OF BLEEDING IN GENGUE INFECTION "– ONE YEAR CROSS SECTIONAL STUDY IN A TERTIARY CARE HOSPITAL"** is a bonafide research work done by, Department of General Medicine, Jawaharlal Nehru Medical College, Nehru Nagar, Belagavi-590010.

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Sub: Acceptance Letter

Sir/Madam,

The softcopy of thesis entitled "STUDY OF PARTIAL THROMBOPLASTIN TIME AND PROTHROMBIN TIME AS A PREDICTOR OF BLEEDING IN DENGUE FEVER – ONE YEAR CROSS SECTIONAL STUDY ", has been submitted for Anti-Plagiarism check through Turnitin software. The scan has been carried out and the scanned output reveals a match percentage of 5% (Five percentage) which is within the acceptable limits of 10% as per the guidelines given by UGC.

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Yours sincerely,

Coordinator
Department of General Medicine
J. N. M. C. Belagavi.

Guide.

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Glossary	Abbreviations
DNV/DEN	Dengue virus
C	Capsid
tPA	Tissue Plasminogen Activator
IL, TNF	Interleukin ,Tumor Necrosis Factor
DIC	Disseminated Intravascular Coagulation
PT	Prothrombin Time
aPTT	Activated Partial Thromboplastin Time
DF	Dengue fever
DF+WS	Dengue Fever with Warning Signs
DHF	Dengue Hemorrhagic Fever
DSS	Dengue Shoock Syndrome
RNA	Ribonucleic Acid
IgG	Immunoglobulin G
IgM	Immunoglobulin M
Pr-M	Membrane Precursor
NS1 antigen	Non-structural protein 1 antigen
E	Envelop
ICAM	Inter Cellular Adhesion Molecule
CD-14	Cluster Differentiation
ER	Endoplasmic Reticulum
SGOT	Serum glutamic oxaloacetic transaminase
SGPT	Serum glutamic-pyruvic transaminase
AT-III	Anti-Thrombin III
USD	United States Dollar
WHO	World Health Organization

ABSTRACT

TITLE : STUDY OF PARTIAL THROMBOPLASTIN TIME AND PROTHROMBIN TIME AS PREDICTOR OF BLEEDING IN DENGUE

INTRODUCTION:

Dengue is a systemic viral infection which is transmitted by mosquitoes such as Aedes aegypti or Aedes albopictus. Dengue fever is characterized by fever , headache, muscle or joint pain and rash. Dengue virus is of 4 subtypes DNV1,DNV2, DNV3, DNV4.All 4 can cause full spectrum of disease from subclinical cases ,mild self limiting infection ,dengue fever ,fatal cases like dengue hemorrhagic fever(DHF) and dengue shock syndrome(DSS).DHF and DSS are related to multiple factors that include thrombocytopenia, coagulopathy and vasculopathy. Coagulopathy results in derangement of activated partial thromboplastin time which is an indicator of impending bleeding risk.

OBJECTIVES:

To measure aPTT levels in dengue virus infection and to identify the role of aPTT in comparison with thrombocytopenia as an indicator of bleeding manifestation to assess the real need of platelet transfusion.

MATERIALS AND METHODS:

A prospective study was conducted from January to august in 2018 in KLE Hospital(JNMC-BELGAVI).Patients with febrile thrombocytopenia who are positive for dengue fever(NS1Ag or IgM) were included in the study. Daily monitoring of platelet count and analysis of aPTT levels were done. aPTT was considered abnormal if it was more than 33 seconds. Patient were followed up for evidence of leaking and bleeding manifestation.

RESULTS:

Out of 100 patients 88 patients had bleeding manifestation. Bleeding signs were seen on clinical examination in 60 patients. Signs of capillary leak was found in the form of pleural effusion in 11 and ascites in 22 patients. Elevated aPTT levels were seen in 43 patients. Among patient with increased aPTT and thrombocytopenia platelet transfusion were done.

CONCLUSION: Our study showed significant correlation between bleeding manifestations and prolonged aPTT levels as well as thrombocytopenia with abnormal aPTT levels. Prolonged aPTT can be used as a predictor of bleeding manifestation especially in DHF and DSS.

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INTRODUCTION

- Dengue fever is a mosquito-borne disease caused by 4 serotypes (DNV1,DNV2, DNV3, DNV4). It is most prevalent arthropod-borne virus affecting human beings**.** Four variants can produce full cycle of disease from subclinical cases which are self-limiting , dengue fever ,fatal cases like dengue hemorrhagic fever(DHF) and dengue shock syndrome(DSS)[2]. Dengue is diffuse throughout the tropics and sub-tropics, with risk factors influencing its occurrence like local climatic variations of rainfall, temperature, relative humidity, the degree of urbanization and quality of vector control services in urban areas. These factors increase the incidence of the disease.[1] Dengue infection is the leading cause of death in tropical and subtropical countries[1]. According to world health organization ,dengue infection is prevalent in more than 125 countries .[2] Worldwide occurrence of estimated dengue infection is around 50-200 million per year. [3,4] First case of dengue fever in India was reported in the year 1780 in "Madras" (now Chennai) and first outbreak occurred in Culcutta (now Kolkata), in the year 1963. Since then, epidemics of dengue have become more frequent in many parts of India mainly in the urban areas.
- Between the years 1998–2009, 82,327 dengue infections (incidence: 6.34 per million population) were recorded. During the year 2010–2014, 213,607 cases (incidence: 34.81 per million population) of dengue fever were recorded. Thus, the number of dengue cases in the past 5 years have rapidly increased. Before 2009, dengue occurrence exceeded 10 per million populations. Since 2010, annual incidence of dengue infection is 15 per million population. [6, 7] .

- The severity of DHF and DSS is determined by multiple factors like, thrombocytopenia , coagulopathy and vasculopathy[3]. Many studies showed a significant correlation between bleeding manifestations in dengue fever and with prolonged aPTT values [2]. Studies have shown that NS1(Nonstructural protein 1) of dengue virus, can bind to both prothrombin and thrombin. NS1 inhibits function of prothrombin by combining with it, and prolongs aPTT values [2]. As there is no specific parameter to predict the bleeding manifestation in dengue fever. Above features can be used to coin new scoring system to find out complications, in early stages of infection. [2].

EPIDEMIOLOGY:

- In Southeast Asia, epidemic of DHF first appeared in the 1950s. By 1975, it had become a leading cause of hospitalization and death among children in many countries. In the 1980s, DHF began a second expansion into Asia [1].
- Different serotypes were observed in 1996 and 2003 outbreaks of dengue fever in north India. DEN-2 and 3 serotypes were identified in these outbreaks respectively. [1]
- Rapid urbanization, inadequate supply of piped water, migration of human population within and between countries have led to the increase incidence of dengue fever .
- There is an increased incidence of insecticide resistance in mosquito vector population. This has led to increase in transmission.[1]

AIMS AND OBJECTIVES

TO STUDY PARTIAL THROMBOPLASTIN TIME AND PROTHROMBIN TIME AS PREDICTORS OF BLEEDING IN DENGUE FEVER – ONE YEAR CROSS SECTIONAL STUDY .

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Dengue virus (DENV) is RNA virus belonging to flaviviridae family. Flavi viruses are passed on to individuals by the bite of arthropod vectors, such as mosquitoes or ticks. Dengue viral infection or "break-bone fever" is a mosquito-borne viral infection, transmitted among humans by the bite of the female mosquito of the *Aedes* genus, of which *A. aegypti* is the main vector, followed by *A. albopictus* , they are distributed mainly in the tropical and subtropical region. The cases of dengue viral infection are influenced by local climatic variation, such as geography of the country, rain-fall, temperature, and rapid urbanization or migration. The outbreak (epidemic) of mosquito-borne infection is a serious issue leading to a rise in number of cases and hyper-endemicity, which can cause a more severe form of dengue infection. Increase in overall spread of dengue epidemic has created public health concern and awareness[15] Prompt history, clinical examination, use of simple diagnostic tests for early detection of disease, use of appropriate treatment protocol and efficient vector control management are the keys, for disease management and control. [15]

STRUCTURE OF DENGUE VIRUS AND ITS REPLICATION:

1. GENOME AND STRUCTURE -

There are four genetically distinct, but antigenically related serotypes classified as DENV-1, DENV-2, DENV-3 and DENV-4 . The dengue virus has a spherical particle existing as either a

50nm diameter immature particle or a mature 60nm diameter particle with a bilayer lipopolysaccharide envelope. The Dengue virus is an enveloped and single-stranded RNA virus. RNA weight is approximately 10.1 kb which is divided into three structural proteins (capsid C, membrane precursor prM and envelop E) and seven non-structural proteins (NS1, NS2A, NS2B, NS3, NS4A, NS4B and NS5)30. The nucleo-capsid is formed by one copy of the RNA genome complexed to multiple copies of the C protein. The nucleocapsid is covered by a double lipid layer in which 180 M proteins and 180 E proteins are anchored. The M protein is formed after the cleavage of the 'pr' portion from the prM protein by a cellular protease (furin) during the process of virus secretion. The glycoprotein E is dimeric and is placed equal to the double lipid layer. [61]

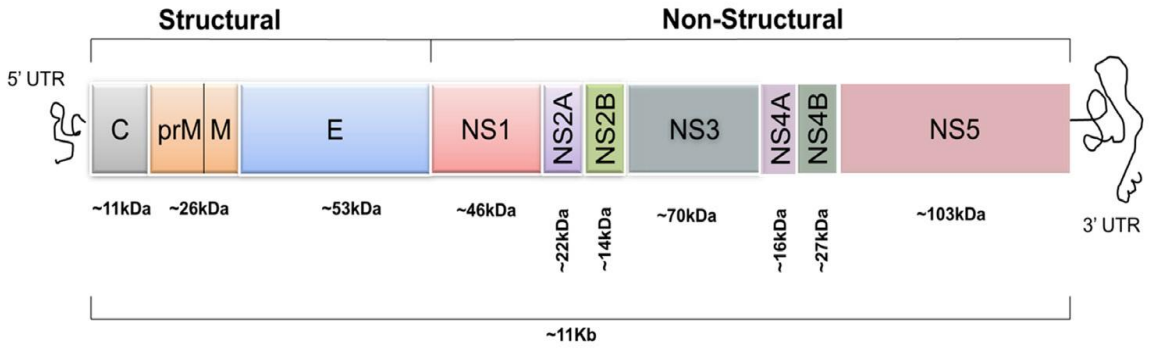


Fig 1.1: Schematic Representation of the Dengue Genome.

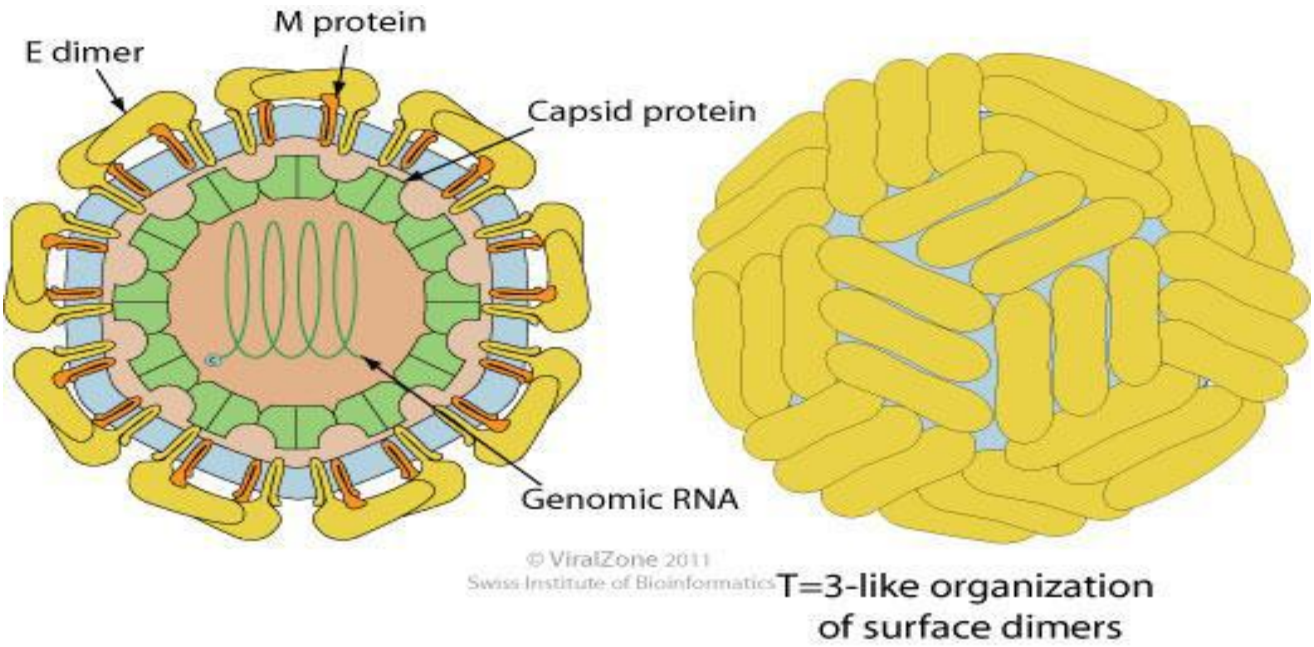


Fig 1.2: Dengue Envelope and Virion Structure.

The immature virus is known as "spiky" as M proteins bound to a "precursor membrane protein" (pr) form heterodimers with E proteins that appear as "spikes" on the viral surfaces. In mature virions the soluble pr is cleaved from M protein by "furin, anchoring the M proteins" and causing the pr protein to be absent in the mature viral membrane.[61]

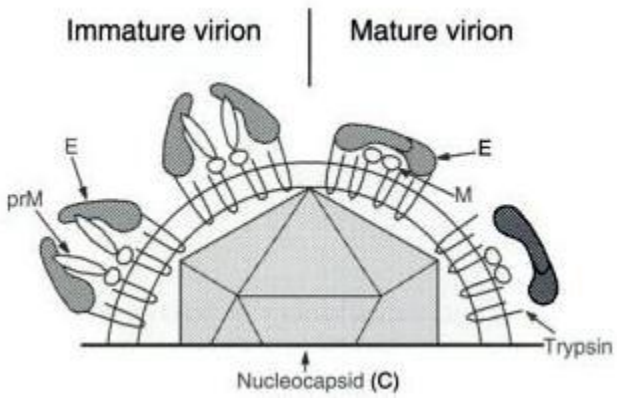


Fig 1.3: Envelope Spikes on Immature and Mature Virions.

2.ENTRY AND REPLICATION -

After a dengue virus infected Aedes. aegypti mosquito feeds on and transfers dengue virons to a human host, the transmitted virus infects susceptible cells through the attachment of the virus to a cell surface receptor. Recognized cell surface receptors are heparin sulfate, a mannose receptor on CD14+ monocytes, and GRP78/Bip on hepatic cells, dendritic cell-specific ICAM3 -grabbing, non-integrin (DC-SIGN) receptor with DENV bound to DC-SIGN. The main mechanism of DENV cell entry is clathrin-mediated endocytosis. However, depending on the serotype and target cell, the virus is able to use alternative internalization routes, consisting of caveolae and lipid rafts. Principal target cells for DENV infection are monocytes, macrophages and dendritic cells. Viral particles spread along the cellular surface and roll over different receptors for the E glycoprotein until they find pre-existing clathrin-coated pits. Clathrin-coated pits evolve until they become clathrin-coated vesicles. These vesicles deliver their cargo to endosomes. It is well established that the mildly acidic pH within the endosomal lumen produces a major conformational change in the E protein, allowing the fusion loop of domain II to be exposed. The highly hydrophobic fusion loop is subsequently inserted into the endosomal target membrane allowing the formation of an E trimer. This E trimer is necessary for the formation of a lipidic fusion pore and release of the genome into the cell cytosol.[61]

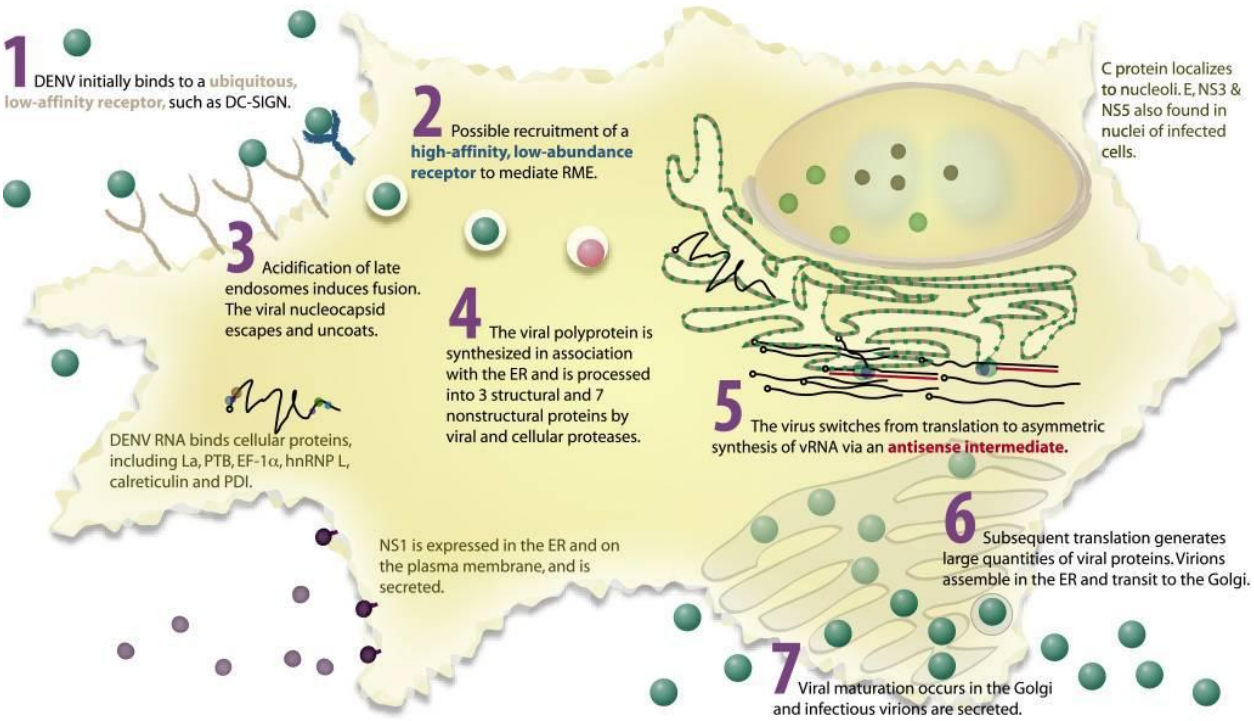


Fig 1.4: Dengue Replication Cycle.

After dengue virions are enclosed in endosomic vesicles they are exposed to acidic pH levels triggering structural modifications of the viral envelope E proteins[61]. This conformational change leads to the exposure of a fusion peptide, which interacts with and bends the endoplasmic membrane towards the viral membrane, inducing the two membranes to fuse. Cellular lysosomal proteases within the endosomic vesicle uncoat the virion and digest the envelope, releasing the dengue genome into the endoplasmic reticulum. Once initiated, ribosomes translate the positive viral RNA strand through the cellular translation machinery to form a polypeptide. This polypeptide is post-translationally cleaved by cellular proteases into structural and non-structural proteins.[61]

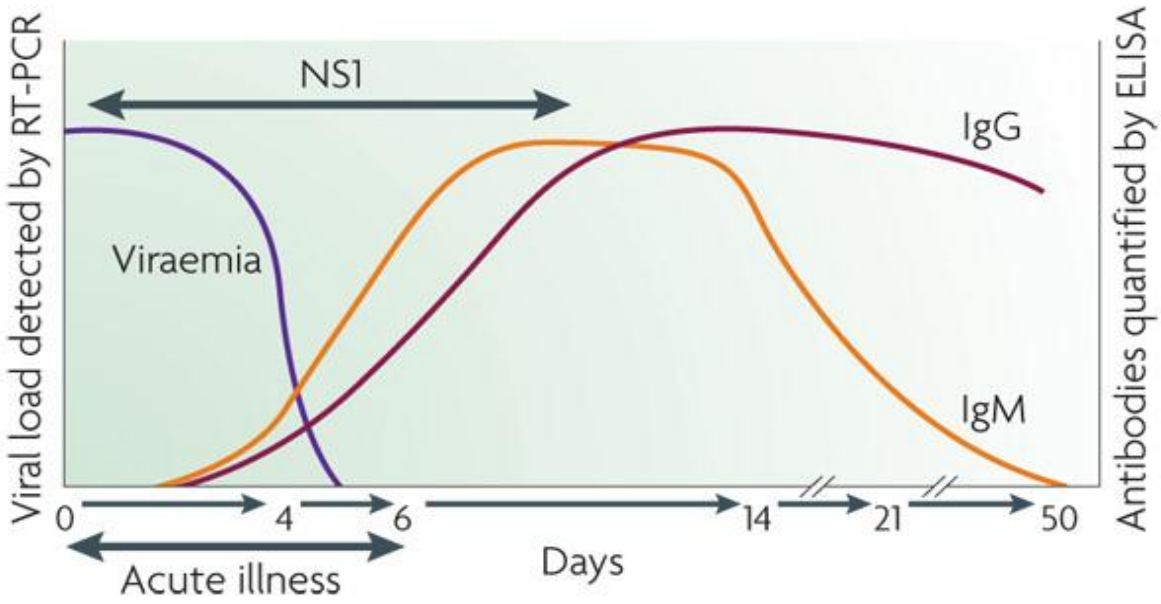
3.Assembly and budding of virus -

On the endoplasmic reticulum of infected cells, DEN Virus is rapidly being packed and assembled into the "virion". The endoplasmic reticulum facilitates the orientation of the capsid, allowing for the association between the viral RNA and the C protein to generate nucleocapsid complexes. The nucleocapsid complex, with the addition of prM and E proteins in the ER, form the heterodimer complex. This complex moves to the golgi apparatus for post-translational modification with the addition of sugar residues. The complex buds through the endoplasmic reticulum lumen, with a prM-E-lipid envelope. While the virus is being exported via exocytic vesicles, mature M proteins form by furin-mediated cleavage of prM and causes E proteins to conformationally change to its homodimer form.[61]

4.Immune Response to Dengue Infection :

Primarly dengue virus infects Langerhans and dentritic cells ,these cells carry the antigen of virus and present it to T-Cells of lymph nodes, which further activates cellular and immune system to kill the virus. Dengue virus specific CD4+ memory T cells and CD8+ T lymphocytes are activated by the T-Cells. Immune system is further activated by production of cytokines and other inflammatory mediators.[61] Activated monocytes and macrophages produces many inflammatory chemokines like tumor necrosis factor,interleukin-1 alpha/1beta/6 and platelet activating factors. These factors, along with complement system and histamine correlates with the complications and severity of illness, due to plasma leakage(increased vascular permeability) leading to shock. First response to infection is by production of antibodies against E and M

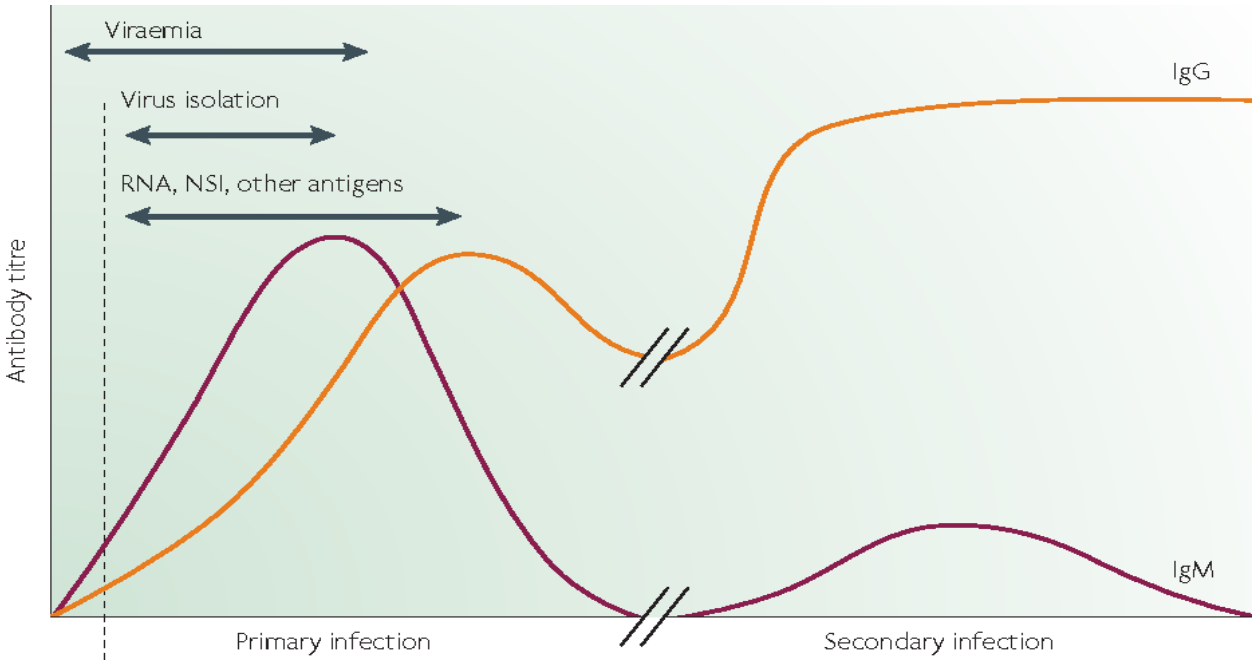
glycoprotein. In the first exposure (known as primary infection) there is slow and low titers of antibodies, as shown in below picture.



NS1 (non-structural protein) is an antigen present over the dengue virus. It is detectable during acute phase of infection. It is positive during first 1-7 days of symptoms. It can be found in whole blood or plasma, most NS1 tests have been developed and evaluated in serum sample. A positive test confirms dengue virus infection without providing serotype information. A negative NS1 test result does not rule out infection. People with negative NS1 result should be tested for the presence of dengue IgM antibodies.[61]

Immunoglobulin M(IgM) antibodies are seen in the early stage. IgM antibodies first seen within 3-10 days levels are maximum in 2 weeks and it decreases over few months. IgG antibodies are detectable in low levels by one week and increases over a time. Each serotype of dengue virus produces unique immunity which is specific to that serotype. Protection can occur after infection

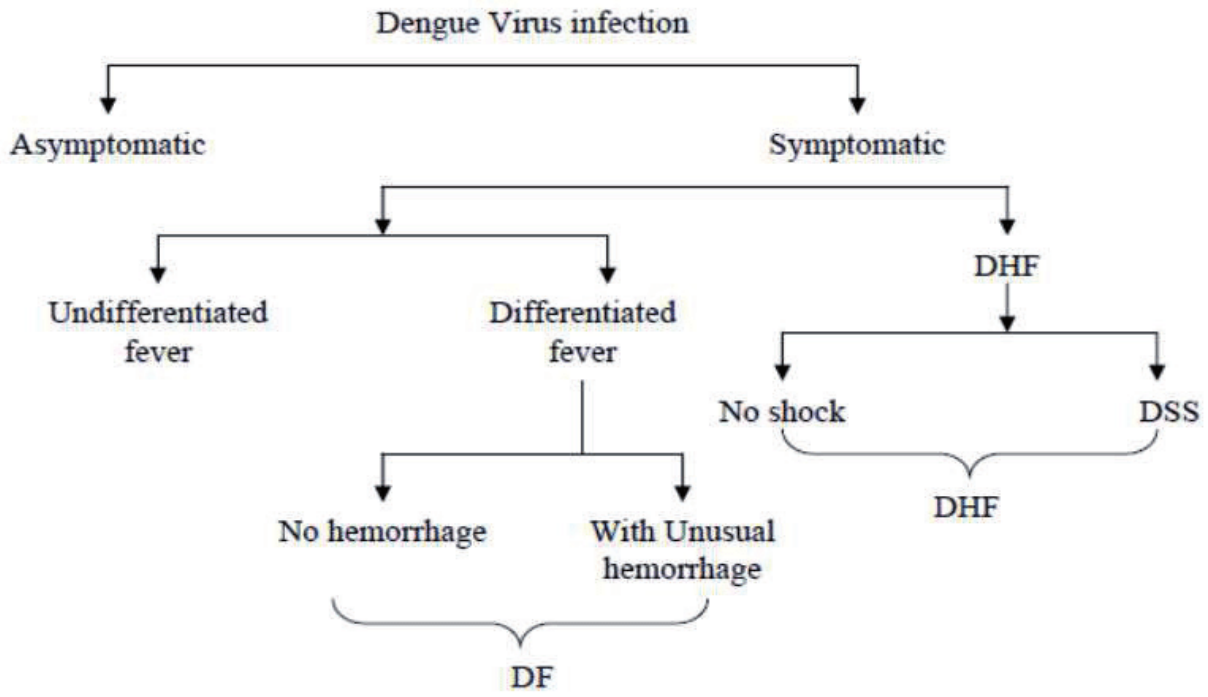
with other serotypes due to cross-protective immunity. During secondary infection(second infection) there are more IgG circulating antibodies and low levels of IgM antibodies. [61]



As shown as above figure.

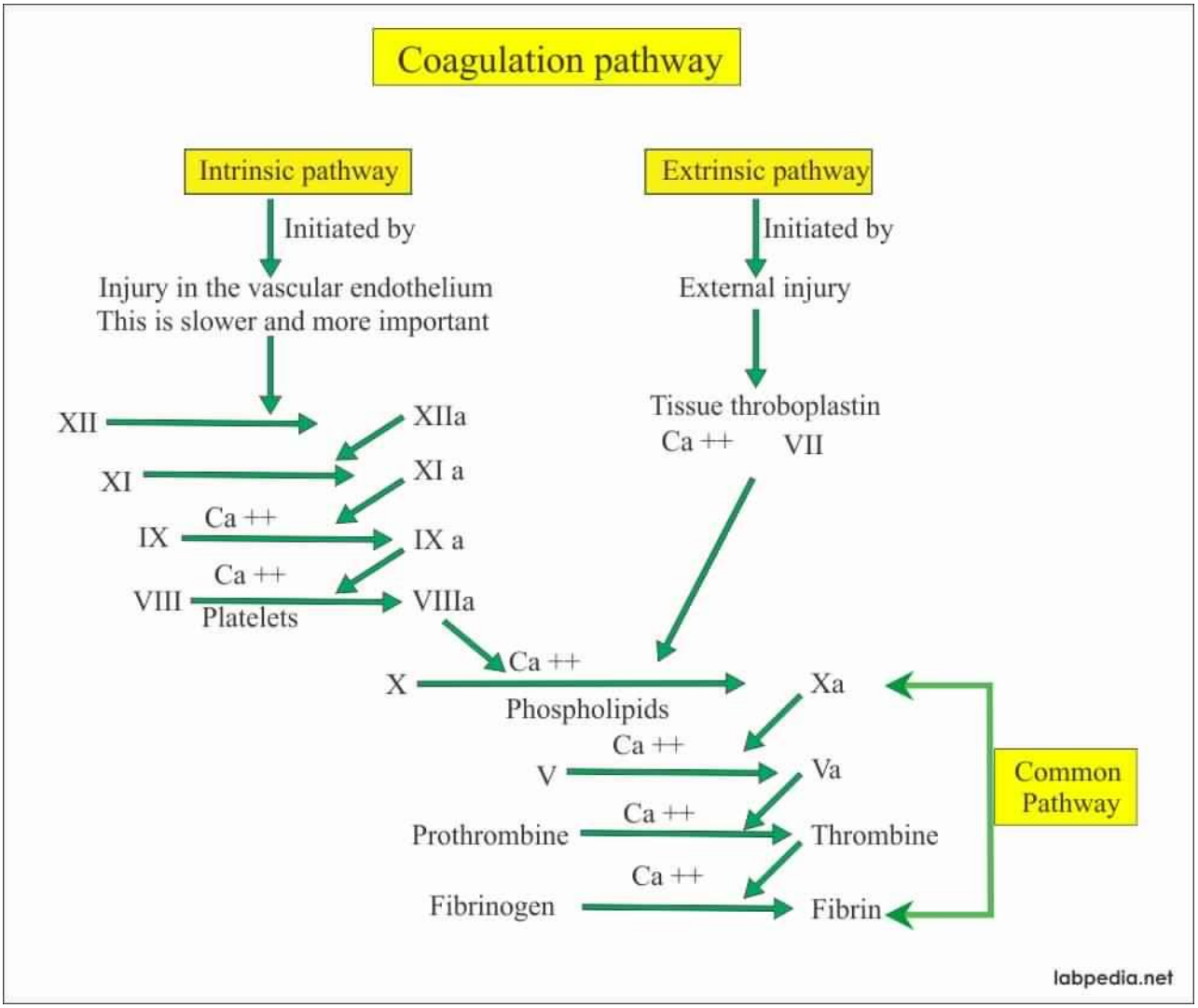
If an individual develops a second dengue infection with heterologous type, these patients are at increased risk of complications related to antibody mediated enhancement(ADE).Due to this process pre-existing antibody recognizes new dengue virus type ,form an antigen-antibody complex ,this complex is non-neutralizing. Virus will be internalized and replicate in the cell which produces more severe infection than primary. [61]

5.Spectrum of dengue infection :



6.Coagulation Pathways:

- Hemostasis is a complex reaction occurring between platelets, coagulation factors, fibrinolytic system, endothelium, pro and anti inflammatory mediators.
- Formation of clot is initiated by insult of vascular system, where platelet plug is formed and it is reinforced with fibrin. [12]
- Anti-thrombin III and activated protein C act as anti-coagulants and can oppose the clot formation.
- Clot formation is balanced by plasma mediated fibrinolysis. [12]



- Armand Quick was the first to demonstrate prothrombin time (PT), which gives information about the extrinsic and common coagulation pathways. PT normal value ranges from 10-13 seconds. High levels of PT are seen in disorders causing low levels of factors II,V, VII, X and fibrinoogen. [12]
- Proctor and Rappaport first demonstrated activated PTT (aPTT) in 1961. Normal aPTT value is 30-40 seconds. It's a clot based test which reflects the intrinsic and common

coagulation pathways. Clinically it is used to monitor the function of unfractionated heparin. [12]

- **RELEVANT ARTICLES :**
- A study was conducted by Abhilash Kannan et al, on coagulopathy in dengue fever patients[10]. In this study , there was a prevalence of coagulopathy in 23% dengue fever patients, as indicated by prolonged aPTT . Results showed abnormality in the intrinsic pathway of coagulation which lasts only for few days during the disease course. PT was normal, which reflected that there was no dysfunction of the extrinsic pathway . aPTT prolongation in the DF patients is due to reduced and impaired synthesis of coagulation factors needed for intrinsic coagulation pathway. Dengue fever is associated with transient coagulopathy during the course of the illness. Apart from thrombocytopenia, coagulopathy also contributes to the bleeding manifestations in Dengue infection . This study showed a direct relation between bleeding tendencies an increased aPTT . In this study they concluded there is a direct relation between thrombocytopenia and raised aPTT. Platelet count decrease is associated with rise in aPTT values.[10]

- A study was conducted by Yng-Huey Huang,et al, on activation of coagulation and fibrinolysis during dengue virus infection [20]. According to this study, hemostatic dysfunctions in dengue infection have multifactorial mechanisms that include, thrombopathy, coagulopathy, and vasculopathy. In this study, PT was normal in all patients with or without hemorrhage , while elevated aPTT was observed in all DHF/DSS

patients. 50% of patients had raised aPTT of 36±45 seconds during the first 2 days after hospitalization [20]. All the patients who had aPTT more than 45 seconds, progressed to hemorrhagic fever or shock syndromes. The above study, concluded there is a direct affinity between severity of disease with increased aPTT[20]. Disseminated intravascular coagulation (DIC), that occurs in DHF and DSS , exhausts both platelets and clotting factors. This may results in prolonged aPTT. [20]

- A study was conducted by Bashir A. et al, on partial thromboplastin time and prothrombin time as predictors for abnormal coagulation among dengue virus infection in Red Sea State of Sudan [25] .They included 336 individuals who fulfilled WHO criteria of dengue fever. Among 336 participants, 106 patients were control. In all diagnosed dengue patients , PT and aPTT were done. Among 336 patients, 35 patients (10.5%) had bleeding symptoms. 5/35 (14.2%) patients had epistaxis, 19/35 (54.3%) had hematuria, and gum bleeding was seen in 9/35(25.7%). 1/35 (2.9%) patients had haematamesis and haemoptysis. Defects in factors like vasculopathy, thrombocytopenia and coagulation system, leads to dysfunction in hemostasis. In this study, they hypothesized that dengue infection activates coagulation system. According to this study NS1(nonstructural protein-1) antigen binds to thrombin and prothrombin[25]. This study, showed that thrombin activity was normal. They concluded that there is prolonged PT and aPTT in dengue infection, which is directly related to impaired coagulation. [25] NS1 inhibits prothrombin which leads to prolonged aPTT. NS1 plays a vital role in vascular leakage. In the initial stages of dengue infection, there is increased release of

inflammatory mediators like, C3a,C5a, TNF-alpha, IL-2,6,10, INF – alpha and histamine. This leads to increase in capillary permeability. [25]

- A study was done by Suchitra Nimmannitya et al, on Dengue Hemorrhagic Fever disorders. They postulated that, hemostatic dysfunction in dengue infection is caused by a multiple factors like, injury to capillaries and venules (vasculopathy) , low platelet count ,abnormal platelet function and dysfunction of coagulation. These abnormal effects are because of cellular dysfunction and activation of immune process by the dengue virus [30]. Main mechanism of vascular dysfunction in DHF is consumptive coagulopathy. Coagulopathy is manifested by abnormally raised aPTT. Thrombocytopenia is due to peripheral utilization and destruction of platelets.[30] Destruction is by activation of complement system, platelets binding to C3g fragments of dengue virus. Abnormal platelet function is also seen. In this study, they observed that there is defect in platelet ADP release and levels of thromboglobulin and platelet factor-4 which are markers of platelet degranulation, are raised in initial phase of dengue infection.[30]

- In a study conducted by Bridget A. Wills et al, on coagulation dysfunction in Dengue Hemorrhagic Fever. They postulated that DSS is consistent with dysfunction of procoagulation, anti-coagulation and fibrinolytic pathways.[35] Cardinal features of DHF is increased vascular permeability. It is associated with hypoalbuminemia, this can be correlated with severity. Anticoagulants like protein C, protein S and antithrombin III are produced in the liver. In DSS, circulating low levels of these anticoagulants hints towards capillary leakage. Capillary leakage is also seen due to complex imbalance between

activated endothelium, platelets and monocytes, followed by dilutional effect of intravenous fluids resuscitation. When there is DIC (in DSS), prothrombin and partial thromboplastin time are raised. [35] Dengue virus directly binds and activates plasminogen and plasmin, which degrade both fibrin and fibrinogen. Hence, bleeding in dengue infection is due to low platelet count, dysfunction of platelets and raised fibrinolysis. [35]

- A study was done by Monroy & B. H. Ruiz on, "Participation of the Dengue Virus in the Fibrinolytic Process".[40] It suggested that, dengue virus has the capability to convert plasminogen to plasmin. They found that, different activation degrees of plasminogen may be associated with different characteristics of virulent strains. Envelope protein of dengue virus has the similar capacity as that of the whole virus activating plasminogen. E surface protein has 3 domains (I,II,III) , domain III of protein has 20% amino-acid homology as anti-plasmin molecule. E surface protein crunches with plasminogen and plasmin and brings conformational changes which can activate or inhibit the enzyme.[40] They proposed that, E protein plays an important factor in maintaining homeostasis. There are high values of PT and aPTT in dengue infection.

- A study was done by Andhika Rachman et al,. on, "Coagulopathy in Dengue Infection"[45] . They saw that raised levels of IL-6 are associated with high auto-antibodies against platelets, tPA (tissue plasminogen activator) levels are raised and coagulation factor XII deficiency, in seen in intrinsic pathway. In acute stage of DHF and DSS, there is high tPA which produces hyperfibrinolysis. They statistically proved the direct association between raised IL-6 and tPA in DHF.[45] Raised levels of IL-6,

down regulates the production of coagulation factor XII. Factor XII is the first factor in intrinsic pathway. Due to deficiency or dysfunction of factor XII there is prolongation of aPTT. Antibodies against E protein can decrease plasmin activity or cause plasminogen activation. There is dysfunction of both coagulation and fibrinolysis which can lead to hemorrhage in DHF and DSS. Dengue virus directly infects the hepatocytes and results in low production of factors for intrinsic pathway. [45]

- A study was done by Yng-huey huang, et al,. on, production of IL-6 and IL-8 in dengue infection. [50] They studied effect of dengue virus on production of cytokines by endothelial cells, these cytokines play an important role in control of hemostasis. They observed that in patients with DHF and DSS, levels of TNF-alpha, IL-6 and IL-8 are raised. Raised IL-6 and 8 can enhance recruitment of leukocytes and can damage to endothelial cells. When an infection with high replication capacity virus occurs , there is sudden and massive release of IL-6 and IL-8 which can lead to DHF and DSS.[50]

- A study was done by Chiou-Feng Lin, et al, on Mechanism of endothelial cell dysfunction in dengue infection. [55] Their study proposed a mechanism where NS1 cross reacts with vascular endothelial cells . This cross reaction causes these cells to undergo apoptosis due to molecular mimicry. Endothelial cells are damaged by Nitric Oxide (NO) . NO causes upregulation of apoptotic process and down regulation of anti-apoptotic factors. [55] There are high levels of IL-6 and IL-8 in patient with DHF and DSS.

- A study was conducted by Whitehorn. J et al,. on, The Pathogenesis and Clinical Management of Dengue [60]. In this study, they tried to understand dengue pathogenesis and risk factors to reduce the morbidity and mortality. They concluded that the genetic differences of MICB(MHC Class I Polypeptide related Sequence B- it's a protein coding gene) and PLCE (Plectin – protein coding gene) genes, can play a role in severity of viraemia in dengue. [60] MICB gene has an important anti-viral function, any mutations are associated with higher vireamia. They found that there was association between severity of dengue infection and genetic variant of these genes.[60] They also found that, both A.aegypti and A.albopictus have similar clinical spectrum in both initial and late dengue infection.

- A study was done by Champika Gamakaranage et al, to find association between dengue hepatitis and abnormal aPTT and PT. Data was collected from 139 patients who were proved cases of dengue infection. [65] They compared the AST levels with aPTT and PT values. They found that, there was no association between high AST levels and aPTT and PT values.[65] Study concluded that, there is no relation between abnormal coagulation parameters in dengue patients.

- Study was conducted by Vijayakumar Balakrishnan et al, on coagulation profile in dengue and its correlation with its severity. [5] They included about 306 proved cases of dengue infection. They monitered PT and aPTT in all patients. There were 56 cases of severe dengue infection in whom 83.9% patients had high PT , INR was > 1.1 in 96.4%

and 91.1% had significantly raised aPTT .[5] In this study, they emphasized the important role of coagulation in dengue infection. Prolongation of aPTT occurs before any immunological variations in dengue. aPTT can be used with other parameters to predict the bleeding manifestation. [5]

- A study was conducted by Kamolwish Laoprasopwattana et al, on PT prolongation as an indicator of severe bleeding manifestation [12]. They found, that PT is affected due to low levels of factor VII of coagulation system. In this study, INR more than 1.5 is a risk factor for development of abnormal liver function test. Prolonged PT is not related to high levels of transaminase[12]. They postulated that normal levels of PT is associated with low rate of bleeding manifestation (it showed a negative predictive value of 89%). Most of the coagulations factors are produced by liver , production is decreased in dengue viral infection which leads to prolonged aPTT , its weakly related with transaminase levels.[12] In this Study, they concluded that gastro-intestinal tract is the most common system to be involved. Platelet count less than 20000 and INR >1.5 is associated with severe dengue viral infection and has more chances of complications.[12].

- According to a review article by Elzinandes Leal de Azeredo et al, on interrelationship between dengue infection and coagulation, fibrinolysis and inflammatory Mediators was studied [22] . They found that low platelet, dysfunction in coagulation and abnormal endothelial cells can lead to hematological manifestation in dengue infection. There is abnormal imbalance between coagulation system and fibrinolysis system. Patients with DSS have abnormalities in both the systems. Factors like low protein C and S is directly

related to increased vascular permeability and related to severity of dengue infection. [22] Antibodies against dengue virus cross react with platelets , endothelial cells and coagulatory factors thus leading to dysfunction in these structures. They concluded that, there are several complex mechanisms involved in the pathogenesis and complications in dengue infection. [22]

METHODOLOGY

STUDY DESIGN:

A ONE YEAR HOSPITAL BASED CROSS SECTIONAL STUDY

SOURCE OF DATA:

This study was Conducted on 100 patients admitted in the wards and ICU of General Medicine department at KLES DR. Prabhakar Kore hospital and Medical Research Centre, J.N.M.C., Belagavi.

STUDY POPULATION :

All the proved cases of dengue fever above age of 18 years, admitted in Medicine department at KLEs DR. Prabhakar Kore hospital and Medical Research Centre, JNMC.

PERIOD OF STUDY:

1STJanuary 2018 to 31STDecember 2018

SAMPLE SIZE:

All the eligible subjects presenting during the study period who satisfied the inclusion and exclusion criteria were included in the study by universal sampling. A total of 100 subjects were enrolled in the study.

Sample size is calculated by the following formula:

N= 4PQ/D2 Where N=Sample size

P = Prevalence of the disease Q= 100- P

D = Absolute error taken as 10%

(P =22.5 [3] ; Q = 77.5 ; D=10)

INCLUSION CRITERIA

1.Confirmed cases of Dengue fever with -

a)WHO clinical criteria :

Including fever, myalgia , retro-orbital pain, severe joint pain, headache,rash and

b)WHO laboratory criteria

-NS1 antigen

-IgM Positive.

2.Patient with thrombocytopenia at the time of Admission(platelet count- less than 150000/mm3) with age above 18 years.

3.Patient with normal packed cell volume values (high PCV Values can interfere with aPTT Values).

EXCLUSION CRITERIA

1.Patient with underlying cirrhosis, Bleeding and Coagulation disorders.

2.Patient with fever and thrombocytopenia who are serologically negative for dengue.

3.Dengue fever with normal platelet count.

4.Patients below 18 years of age.

Ethical considerations: Study was approved by the " institutional human ethics committee".

Informed written consent was obtained from all the study participants and only those

participants willing to sign the informed consent were included in the study. The risks and benefits involved in the study and voluntary nature of participation were explained to the participants, before obtaining consent. Confidentiality of the study participants was maintained.

Data collection tools: All the relevant parameters were documented in a structured study proforma.

Methodology:

Patients fulfilling the above inclusion and exclusion criteria were enrolled in the study, after obtaining written informed consent. History, clinical examination and investigations were documented. A probable case of dengue infection was diagnosed according to the WHO criteria:

Dengue Fever:

Acute febrile illness with two or more of the following:

- A headache,
- Retro-orbital pain,
- Myalgia,
- Arthralgia,
- Rash,
- Hemorrhagic manifestation,
- Leucopenia (WBC<5000 cells/mm),
- Thrombocytopenia –platelet< 150,000 cells/mm,
- Rising hematocrit (5-10%)

Dengue Haemorrhagic Fever:

- All of the above with following:
- Hemorrhagic manifestation, shown by any of the following: positive tourniquet test, petechiae, ecchymosis or purpura, or bleeding from mucosa, gastrointestinal tract, injection site.
- Platelet count < 100,000 cells/mm
- Rising hematocrit > 20% from baseline

Dengue Shock Syndrome:

- Criteria for dengue hemorrhagic fever as above clinical findings with signs of shock including:
- Tachycardia, cool extremities, delayed capillary refill, weak pulse, lethargy
- Pulse pressure <20 mmHg with increased diastolic pressure.
- Hypotension by age, defined as systolic pressure < 90 mmHg.

Investigations:

- CBC
- NS1 Antigen
- Dengue IgM antibody
- Liver function tests
- Kidney function tests
- PT and Aptt.

RESULTS

Methods used: Data analysis was done using R i386 3.5.1.Data is summarised as mean±SD for continuous variable and categorical variables are represented using percentages. Comparison of aPTT and PT times between 2 serological tets are done using independent t-test / Mann-Whitney U test and categorical variable between 2 serological (NS1 and IgM) tests are compared using fisher test. P-value <0.05 is considered as significant.

100 subjects of age group 31.81±12.50 years (range: 18-75) were enrolled for the study, consisting of 74 male and 26 female subjects. summary of data shown in table 1.

Table 1: Descriptive statistics of Age in the sample.

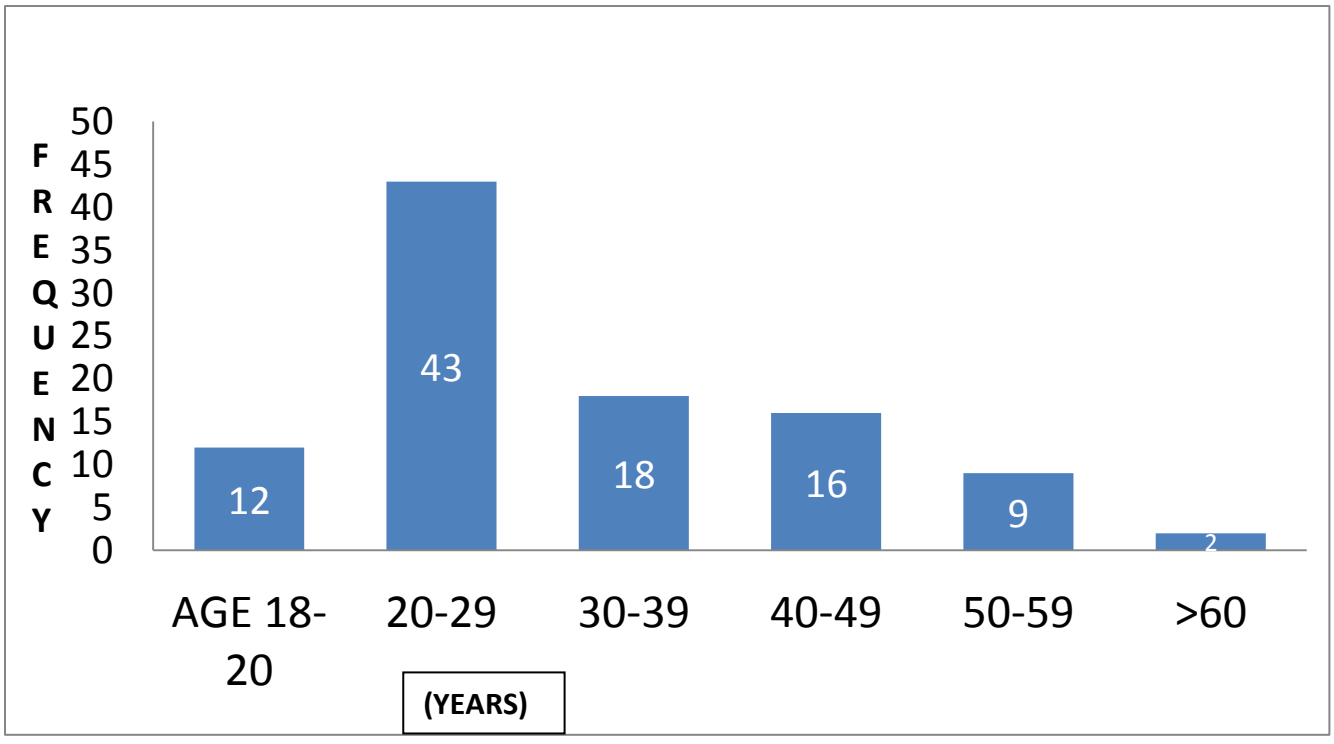
Factor	mean±sd	median	minimum	maximum	95% CI	
					Lower	upper
Age	31.81±12.50	28.5	18	75	29.32	34.29

From table 1, we observe that Mean age of subjects is found to be 31.81 ± 12.50(years). The minimum age of subjects is 18 years and maximum age is found to be 75 years.

Table 2: Distribution of subjects over Age group in the sample.

Age group	Frequency	Percentage (%)
18-20	12	12%
20-29	43	43%
30-39	18	18%
40-49	16	16%
50-59	9	9%
>59	2	2%

Figure 1: Age group wise distribution of subjects in the Sample.

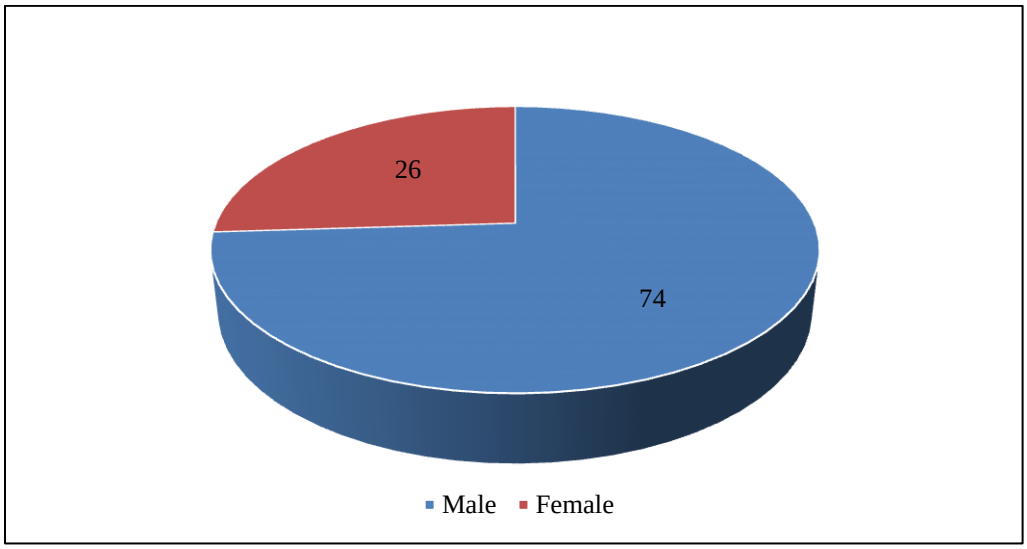


From table 2and Fig-1, we observe that Among the total subjects, majority (43%) of the subjects in the sample are in the age group 20-29 years followed by 30-39 years (18%) and 16(16%) subjects are in between the age group 40-49 years.

Table 3: distribution of subjects by gender in the Sample.

Gender (N= 100)	Frequency	Percentage (%)
Male	74	74%
Female	26	26%

Figure 2: distribution of subjects by gender in the Sample.



From table 3 and Fig-2, we observe that the sample data consist of 74 (74%) male subjects and remaining 26(26%) are female subjects.

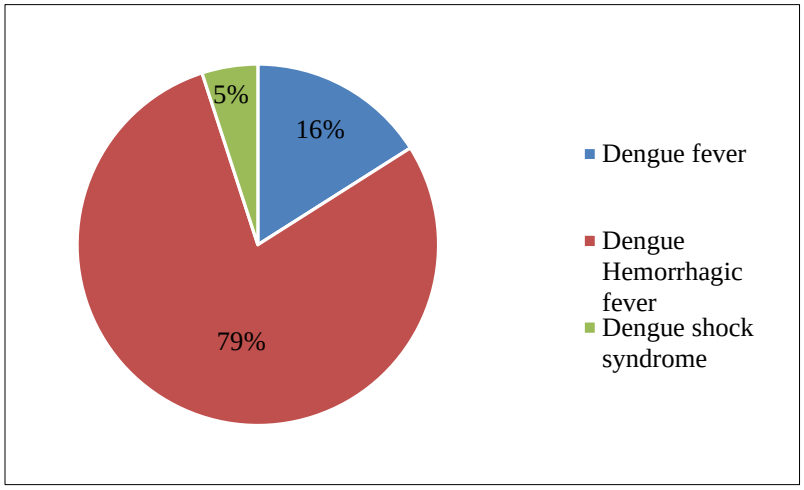
TABLE 4 : Distribution of Subjects according to clinical diagnosis.

Factor		Frequency	Percentage (%)
Diagnosis (N= 100)	**DF**	16	16%
	DHF	79	79%
	DSS	5	5%

(DF: Dengue fever; DHF: Dengue Hemorrhagic fever; DSS:Dengue shock syndrome)

(Table 4 showing 79% of the total subjects were diagnosed with the Dengue Hemorrhagic fever followed by Dengue fever (16%) , 5% were diagnosed as Dengue Shock Syndrome)

Figure 3: : Distribution of Subjects according to clinical diagnosis in the Sample.



(DF: Dengue fever; DHF: Dengue Hemorrhagic fever; DSS:Dengue shock syndrome)

(Graph:3 showing 79% of the total subjects diagnosed with the Dengue Hemorrhagic fever followed by Dengue fever (16%) , 5% diagnosed as Dengue Shock Syndrome)

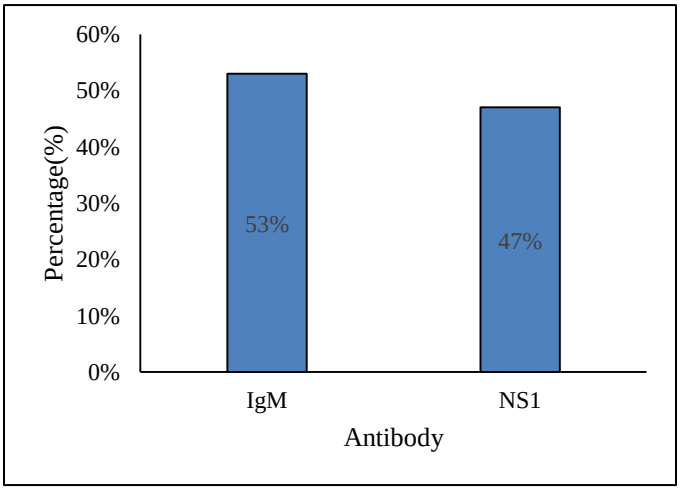
Table-5: Dengue test interpretation in subjects (n=100).

Factor		Frequency	Percentage (%)
Dengue Profile (N= 100)	**IgM**	53	53%
	NS1	47	47%

Abreviation: IgM: Immunoglobulin M; NS1: Non-structural protein 1

Table: 5 showing 53% of the subjects had dengue IgM antibody positive and 47% of the subjects were NS1 antigen positive)

Figure 4: Bar Graph showing interpretation of Dengue test.



(Abreviation: IgM: Immunoglobulin M; NS1: Non-structural protein 1)

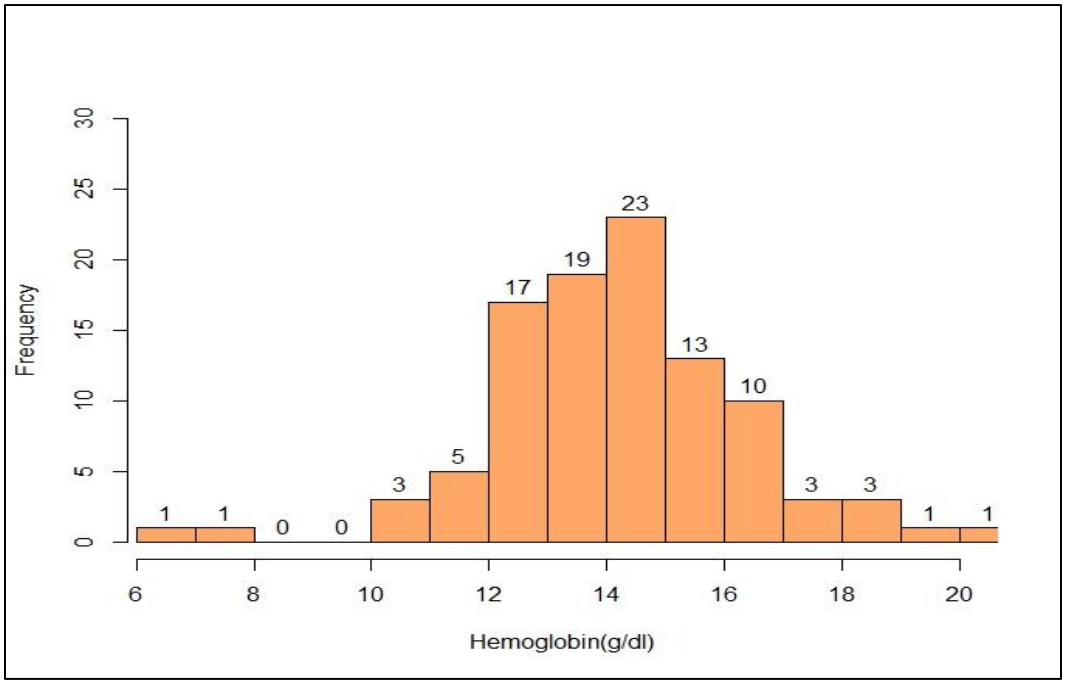
. (**Graph:4** Showing 53% of the subjects as IgM antibody positive while 47% of the subjects showing NS1 antigen positive)

Table 6: Descriptive statistics of Hemoglobin in the Sample

Factor	mean±sd	median	minimum	maximum	95% CI	
					Lower	Upper
Hemoglobin(N= 100)	14.26±2.22	14.2	6.4	20.1	13.82	14.70

From table 6, we observe that Mean hemoglobin of sample is 14.26 ± 2.22 (g/dl) and is ranged between 6.4 - 20.1 (g/dl)

Figure 5: Histogram of Hemoglobin Distribution in the Sample.



From the above figure-5 , we observe that, majority (23%) of the total subjects have hemoglobin range between 14-15 (g/dl) followed by 13-14(g/dl).

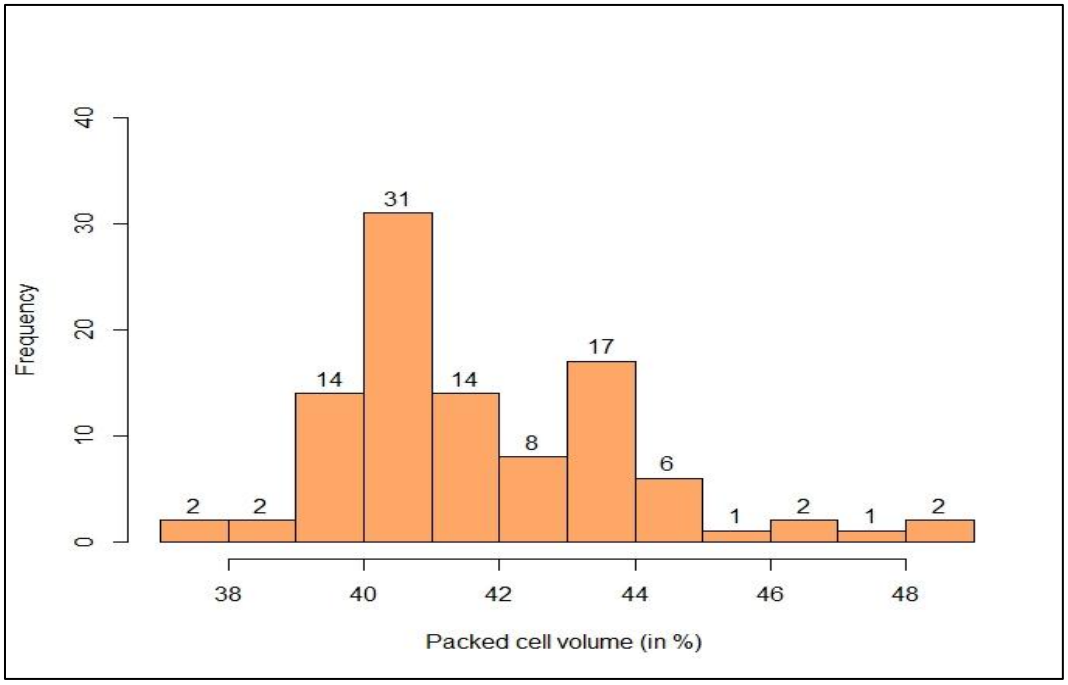
Table 7: Descriptive statistics of packed cell volume in the Sample

Factor	mean±sd	median	Minimum	maximum	95% CI	
					Lower	Upper
packed cell volume (PCV) (N= 100)	42.02±2.11	41.4	37	48.9	41.60	42.43

.

From Table 7, we observe that Mean PCV of the sample is 42.02 ± 2.11 (%) .The maximum of PCV is 48.9% whereas the minimum of PCV in sample is 37%.

Figure 6: Graph Showing distribution of PCV(packed cell volume) in the Sample.



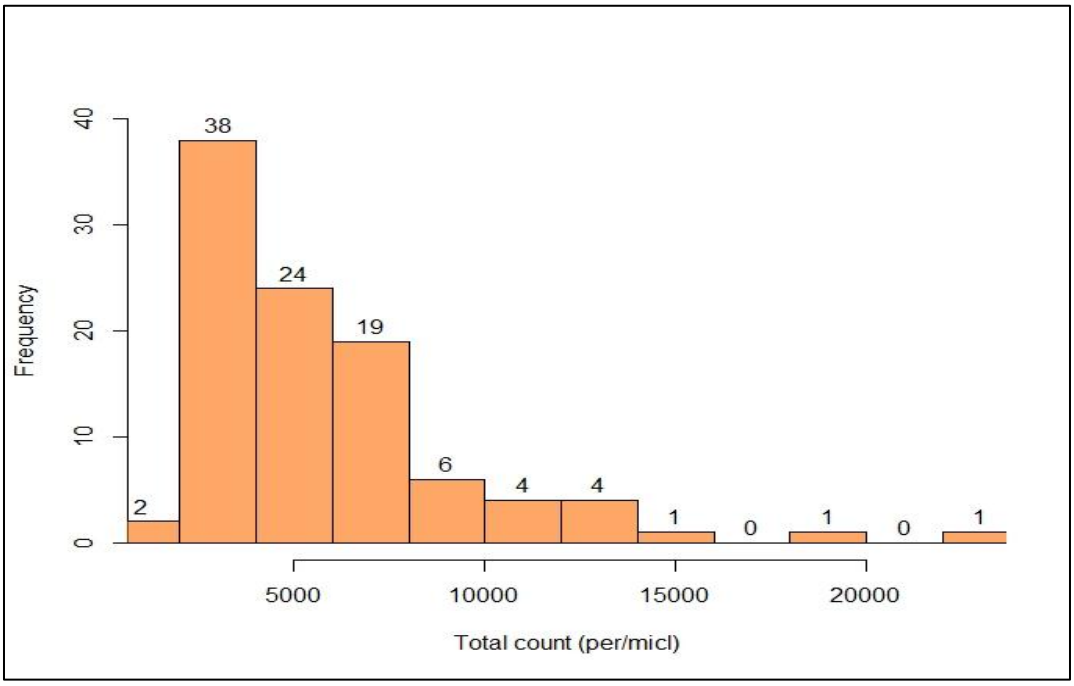
From graph:6 shows, around 31% of the total subjects had the PCV between 40% to 41% followed by 43%-44% (17%).

Table 8: Descriptive statistics of White Blood Cell Count in the Sample.

Factor	mean±sd	median	minimum	maximum	95% CI	
					Lower	Upper
Total count(N= 100)	5701±3575.01	4700	1600	22800	4991.64	6410.36

From the table 8, we observe that the range of total count in the sample is in between 1600 – 22800 with Average 5701 ± 3575.01 (per/micl).

Figure 7: distribution of White Blood Cell count in the sample.



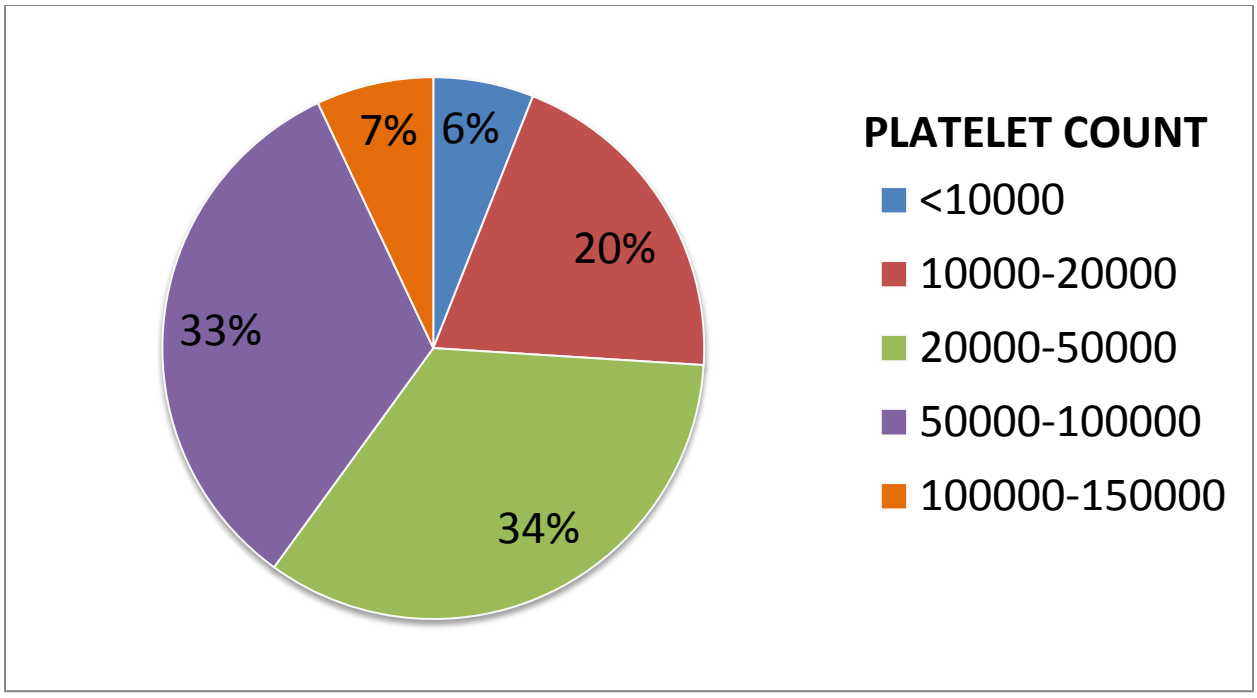
From figure 7, we observe that majority of the subjects had total count in between 2000 to 4000 followed by 4000-6000 (24%).

Table 9: Descriptive statistics of Platelet Count in the Sample.

Factor	mean±sd	median	minimum	maximum	95% CI	
					Lower	Upper
Platelet count (N= 100)	494.10±366.95	380	200	1490	421.29	566.91

From table 9, Mean platelet count of the sample is 494.10 ± 366.95 / cumm with a range of 200 - 1490 /cumm.

Figure 8: Distribution of Platelet count in the sample.



From the figure 8, Above graph shows 6 patient had platelet below 10000/cumm, 20 patients had platelet count 10000-20000/cumm, 34 patients had platelet between 20000-50000/cumm, 33 patients had platelet count between 50000-100000/cumm and 7 patients had platelet count between 100000-150000/cumm.

Table 10: Descriptive statistics of prothrombin time and aPTT in the Sample.

Factor(N=100)	mean±sd	median	minimum	maximum	95% CI	
					Lower	Upper
Prothrombin time(seconds)	12.29±1.90	11.6	10.1	19.7	11.91	12.67
APTT(seconds)	39.25±7.22	38.45	20.8	58.5	37.82	40.68

Abreviation: APTT: activated partial thromboplastin time

From table 10, we observe that range of Prothrombin time in the sample is (10.1 - 19.7 sec) with mean time 12.29±1.90. Mean aPTT is found to be 39.25 ± 7.22(sec) with a range between 20.8 sec and 58.5 sec.

Figure 9: graphical representation of PT and Aptt in the Sample.

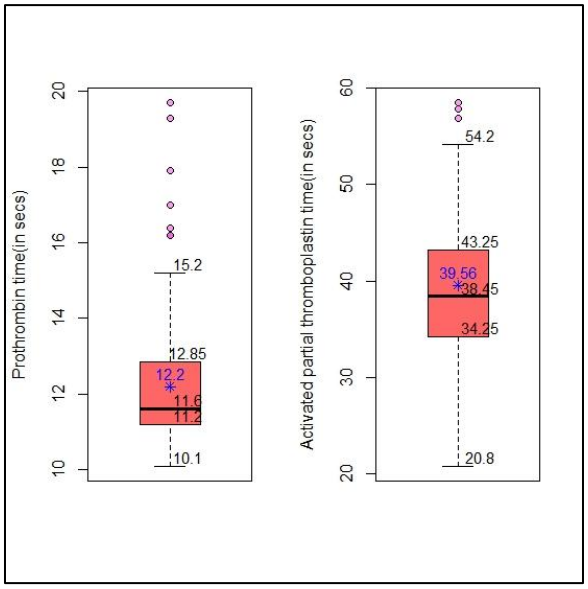


Fig.9 showing ranges of PT and aPTT.PT ranges from 10.1 to 12.85 seconds and aPPT ranges from 34.25-43.25 seconds.

Table 11: comparison of different of PT and aPTT values between NS1 antigen and IgM antibody. .

Factor	Dengue Test		P-value
	NS1	IgM	
Prothrombin time (seconds)	11.6 (11.2-12.8)	11.6(11.2-13.05)	0.6986
aPTT (seconds)	38.5(34.9-43.2)	37.9(34.15-43.6)	0.7507

From table 11, using Mann-Whitney U-test, we conclude that there is no significant difference in median value of Prothrombin time as well as activated partial thromboplastin time between NS1 and IgM patients. Using fisher test, we conclude that, there is no significant association between Diagnose and serology of dengue. From figure 9, we visualize the results.

Table 12: Showing Distribution of the Spectrum of Dengue infection according the Dengue Profile.

Factor		Dengue Test		P-value
		NS1	IgM	
Diagnose	**DF**	10(21.28)	6(11.32)	0.4324
	DHF	35(74.47)	44(83.02)	
	DSS	2(4.25)	3(5.66)	

Table:12-In NS1 positive patients, 10 had Dengue Fever, 35 had Dengue Hemorrhagic Fever and 2 had Dengue Shock Syndrome. In IgM antibody positive patients, 6 had Dengue Fever,44 Dengue Hemorrhagic Fever and 3 patients had Dengue shock syndrome.

Figure 10: Comparison of different Prothrombin Time and aPTT between NS1 and IgM.

9(a) 9(b)

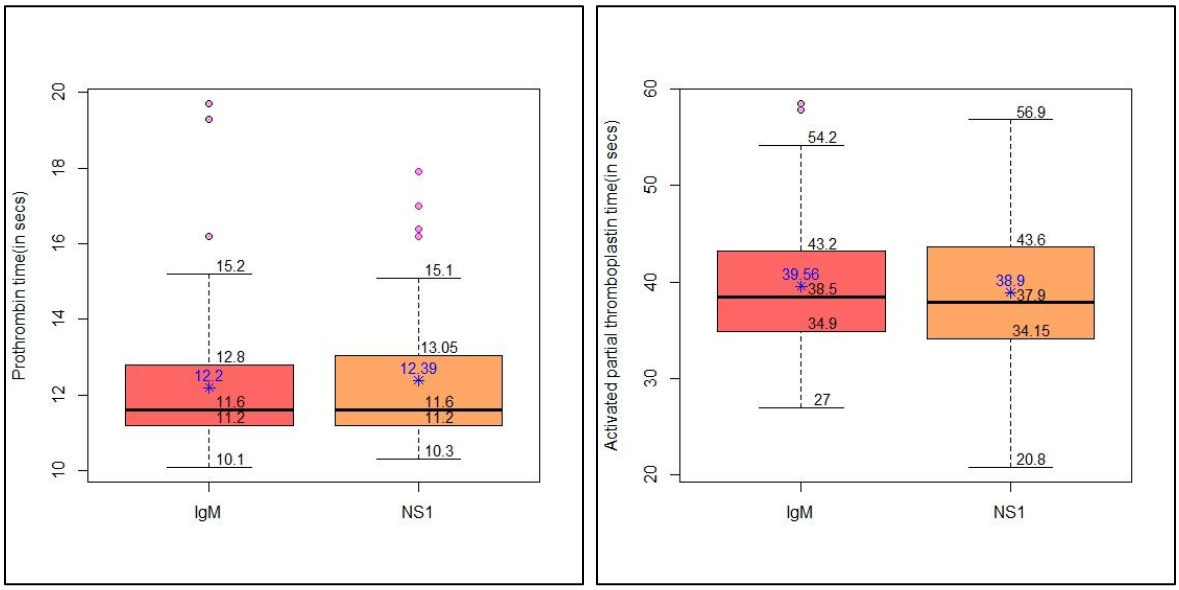
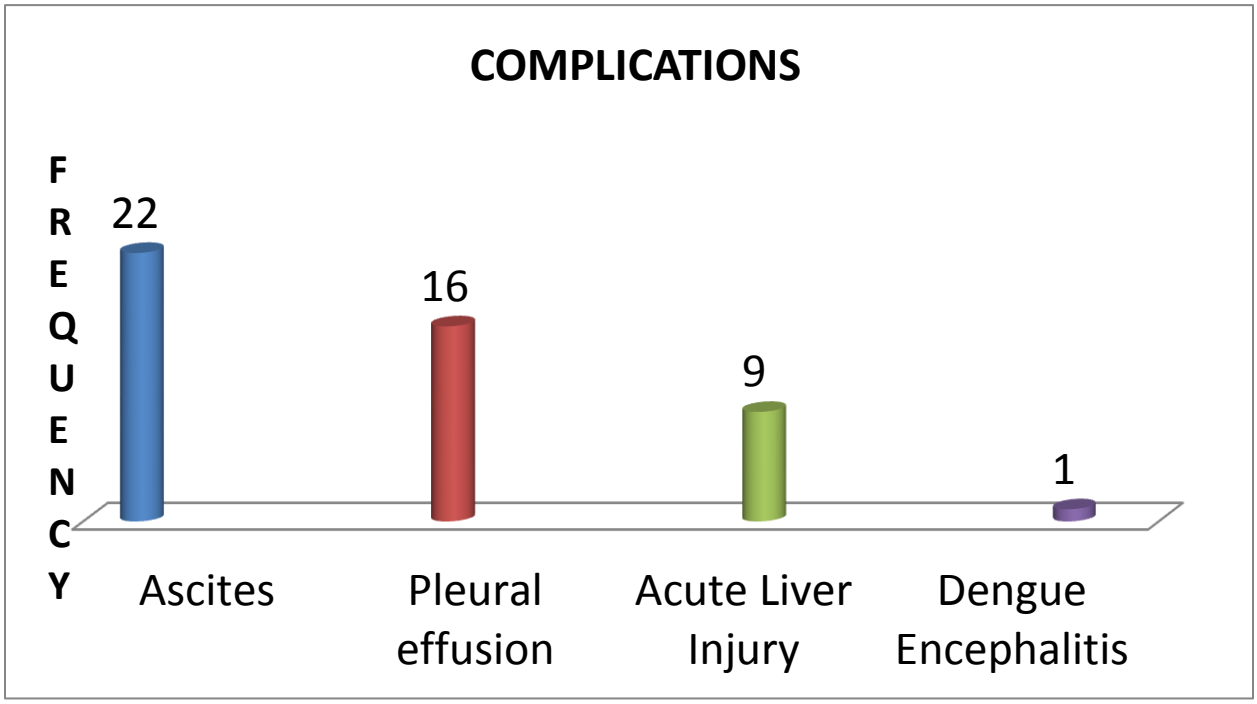


Table 13: Association between PT as well as aPTT category with diagnosis

Factor	Levels	Diagnose			P-value
		DF	DHF	DSS	
PT category (seconds)	**11-15**	15(93.75)	72(91.14)	4(80)	0.5977
	15-20	1(6.25)	7(8.86)	1(20)	
APTT Category (seconds)	**28-35**	12(75)	21(26.58)	0	0.0012*
	35-40	3(18.75)	27(34.18)	1(20)	
	40-45	1(6.25)	13(16.46)	1(20)	
	>45	0	18(22.78)	3(60)	

.From table 13, using fisher test, we conclude that there is no significant association between Pt category and diagnosis while there is a significant association between aPTT category and diagnosis.

Figure-11:The below graph shows complications of dengue fever observed in this study.



Above graph shows 22 patients had ascites, 11 patients had pleural effusion , 9 patients had acute liver injury and 1 patient had dengue encephalitis

Table-14- Table showing co-relation between complications and platelet count, PT, aPTT .

COMPLICATIONS (N=43)	PLATELET COUNT (/cumml) (Average)	PROTHROMBIN TIME(SECONDS) (Average)	ACTIVATED PARTIAL THROMBOPLASTIN (SECONDS) (Average)
ASCITES (N=22)	14,645	11.30	42.58
PLEURAL EFFUSION (N=11)	15,636	12.03	45.70
ACUTE LIVER INJURY(N=9)	28,000	14.03	43.21
DENGUE ENCEPHALITIS (N=1)	6000	11.4	44.7

Above table shows Complications in this study. Ascites is the most common complication followed by pleural effusion, acute liver injury and Dengue encephalitis. Platelet count was below 20000/cumm in all complications except in acute liver injury. aPTT was raised in all complications. PT was raised mainly in acute liver injury.

DISCUSSION

Dengue infection is the most prevalent mosquito related viral infection worldwide. About 50 million patients are diagnosed with dengue fever each year, out of which 5 lakh individuals are admitted. [64] Dengue infection is distributed in the regions of Pacific, America and Southeast Asia. In India it is one of most common mosquito-vector born disease. It has an economic burden of 27.3 million united states dollars · Previously dengue infection was mainly distributed in the northern states namely Haryana, Punjab and Uttar Pradesh, now it has assumed pan–India distribution. It is widely seen in states of Andhra Pradesh, Tamil Nadu and Karnataka, Gujarat , Rajasthan; and west Bengal. Dengue infection varies from an asymptomatic to a symptomatic spectrum. There is no specific treatment protocol. Main aim of treatment is fluid management and careful monitoring of vital signs and correction of metabolic derangements. Dengue infection damages the hepatocytes and decreases the production of coagulation factors needed for coagulation system activation(mainly intrinsic pathway). The abnormal coagulation profile , reflected by raised PT and aPTT duration can be used to predict the complications of dengue infection. [64]

In this study ,we included 100 dengue infection patients as per the WHO guidelines.

AGE AND GENDER DISTRIBUTION:

We included patients above 18 years of age. The average age group was 28.5 years.

In this study , 74% are males and 26 % are females.

This was in concordance with a similar study conducted by Bashir et.al. Their Study included a total of 334 patients, with age ranging between 6-76 years(mean 22 years) . There were 217(65%) males and 117(35%) females. [25]

Study done by Champika Gamakaranage et.al , included 137 patients ranging from 12-82 years ,with a mean age of 33 years. There were 71 males and 66 females(M:F = 1.167). [65]

Diagnosis of Dengue fever:

Out of 100 cases of dengue infection in our study, 53 cases are NS1 positive and 47 cases are IgM Positive.

Platelet count :

Out of total 100 patients, 6 patients had platelet count below 10000/cumm , 20 patients had platelet count 10000-20000/cumm, 34 patients had platelet between 20000-50000/cumm, 33 patients had platelet count between 50000-100000/cumm and 7 patients had platelet count between 100000-150000/cumm. About 93 patients had platelet count less than 100000/cumm.

Abnormal PT/aPTT :

In our study, we have compared the high PT and aPTT values with the spectrum of the disease. In NS1 positive patients, prothrombin time ranges from 11.2-12.8 seconds and aPTT ranges from 34.9-43.2 seconds.

In IgM positive patients, PT ranges from 11.2-13.05 seconds and aPTT ranges from 34.15-43.6 seconds. In both NS1 and IgM positive patients, aPTT was raised more than PT in both dengue hemorrhagic fever(DHF) and dengue shock syndrome(DSS). Hence, aPTT can be used to predict the severity in dengue infection.

This is in concordance with a similar study done by kannan A et al.,(2014). In their study, they categorised aPTT values into 2 categories namely, normal (<40 seconds) and abnormal (>41

seconds).They found a significant ‘p’ value and a strong co-relation between raised aPTT and severity of dengue. They compared platelet count and aPTT values , they observed that as the aPTT value rises, the bleeding manifestations also increased.

A similar study was conducted by A. Bashir et al., which included 334 patients . Patients were screened with PT and aPTT. The median aPTT was 32.4seconds(mean ± SD 33.5 ± 9.7) (range 15.0 – 80.7seconds) and PT 13.8 seconds (mean ± SD 14.1 ± 2.1) (range 10.0 – 20.4 seconds). PT and aPTT were found to be relatively higher in DF(90% and 72.7%) and DHF(10% and 27.3%). They concluded that raised PT and aPTT is associated with bleeding manifestations. PT and aPTT can be used as predictor for bleeding in dengue infection.

A study conducted by Champika Gamakaranage et al.,(2018) , included 139 patients of dengue fever. They tried to establish a relation between raised aspartate and altered coagulation profile. They found negative co-relation, but a high PT and aPTT was associated with bleeding manifestations.

BLEEDING MANIFESTATION :

In our study, the commonest bleeding manifestation noted was malena and skin petechie. Other bleeding manifestations were conjunctival congestion , epistaxsis and bleeding gums .The other complications were pleural effusion and acute liver injury. One patient developed dengue encephalitis.

Complication :

In this study 22, patients had ascites, 11 patients had pleural effusion , 9 patients had acute liver injury and 1 patient of dengue encephalitis . aPTT was raised in all complications but PT was increased only in patients with acute liver injury. All patients with complications had platelet count less than 30,000/cumm.

Other findings:

1. We observed that, majority (23%) of the total subjects had hemoglobin in the range of 14-15 g/dl followed by 13-14 g/dl**.**

2. We observed that mean PCV of patients was 42.02 ± 2.11 (%) .The maximum PCV was 48.9% whereas the minimum of PCV was 37%. Around 31% of the total subjects had PCV between 40% to 41% followed by 43%-44%(17%).

3. We observed that the range of total white cell count in this sample was between 1600 – 22800 , average 5701 ± 3575.01 (per/cum). Majority of the subjects had total count in between 2000 to 4000 followed by 4000-6000 (24%).

4. Majority of the subjects had platelet count less than 40000/cumm. while 18% of the subjects had platelet count between 90000 to 100000/cumm.

CONCLUSION

- In our study, the average age was between 31.81±12.50 years (range: 18-75 years).

- In the present study, 74% were male and 26% were female subjects.

- In our study, 53% of the subjects had IgM antibody positive while 47% of the subjects had NS1 antigen positive.

- 79% of the total subjects were diagnosed with Dengue Hemorrhagic fever followed by Dengue fever (16%).

- We observed that, mean hemoglobin in our study was 14.26 ± 2.22 (g/dl). Majority (23%) of the total subjects had hemoglobin between 14-15 (g/dl) followed by 13-14(g/dl).

- We observed that, mean PCV was 42.02 ± 2.11 (%) .The maximum PCV was 48.9% whereas the minimum PCV was 37%. Around 31% of the total subjects had PCV between 40% to 41% followed by 43%-44% (17%).

- We observed that majority of the subjects had total white cell count in between 2000 to 4000cells/cuml followed by 4000-6000 (24%).

- We observed that, 6 patient had platelet below 10000/cumm,20 patients had platelet between 10000-20000/cumm, 34 patients had platelet between 20000-50000/cumm, 33 patients had platelet count between 50000-100000/cumm and 7 patients had platelet count between 100000-150000/cumm .

- We observed that the range of prothrombin time in our study was (10.1 - 19.7 sec) with mean time 12.29±1.90 seconds. Also, mean aPTT was found to be 39.25 ± 7.22(sec) with a range between 20.8 sec and 58.5 sec.

- We observed that , ascites is the most common complication in this study followed by pleural effusion ,acute liver injury and Dengue encephalitis. Platelet count was below 20000/cumm in all complications except in acute liver injury. aPTT was raised in all complications. PT was raised mainly in acute liver injury.

  .

- Our findings revealed that there was a significant positive correlation between raised aPTT levels with severity of dengue fever. PT is raised in acute liver injury.

- We concluded that, there was no significant difference in median value of prothrombin time as well as activated partial thromboplastin time between NS1 positive and IgM positive patients.

- We concluded that there is direct co-relation between bleeding manifestation and raised aPTT.

LIMITATIONS

- Analysis to identify independent predictors of raised aPTT and PT values among the study population was not possible as the sample size of study was limited.

- The generalizability of study findings was limited, as the study was done on a small sample of the population attending a tertiary hospital.

SUMMARY:

In this study we included 100 patients who fulfilled the WHO criteria of dengue fever. 74 patients were males and 26 patients were female. Most of the patients had dengue hemorrhagic fever. Majority of the patients had platelet count less than 50000/cumml. We observed, that there was increased aPPT in patients with complications. Most common complications in this study was ascites followed by pleural effusion. We observed there is direct co-relation between raised aPPT and complications.

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ANNEXURES

PROFORMA

THESIS TOPIC- PARTIAL THROMBOPLASTIN TIME AND PROTHROMBIN TIME AS PREDICTORS OF BLEEDING IN DENGUE FEVER

CASE NO:

NAME:

AGE/SEX:

IP NO :

COMPLAINTS AT PRESENTATION:

PHYSICAL EXAMINATION:

PALLOR- YES/NO

ICTERUS-YES/NO

LYMPHADENOPATHY-YES/NO

CYANOSIS- YES/NO

CLUBBING-YES/NO

EDEMA-YES/NO

VITALS:

TEMPERATURE :

PULSE :

RESPIRATORY RATE :

BLOOD PRESSURE :

DEHYDRATION - YES/NO

SYSTEMIC EXAMINATION :

CVS -

RS -

P/A -

CNS –

SIGNS OF BLEEDING MANIFESTATION:

- PETECHIE - YES/NO
- MALENA - YES/NO
- CONJUNCTIVAL CONGESTION - YES/NO
- HEMATEMESIS - YES/NO
- HEMOPTYSIS - YES/NO
- OTHERS -

INVESTIGATION :

1.Dengue profile -

NS1	
IgM	

2. Platalet count –

3.Coagulation profile -

	PT	aPTT
Control		
Test		

4.Liver Function Test –

T.Bili / D.Bili	SGOT / SGPT	T.Protein / Albumin	AKLPh

5.Renal Function Test:

Urea	S.Creatinine	Na	K	CL

COMPLICATIONS :

DIAGNOSIS :

- Dengue fever (DF)
- Dengue hemorrhagic fever (DHF)
- Dengue shock syndrome (DSS)

INFORMED CONSENT

Title Of Research Study:TO STUDY PARTIAL THROMBOPLASTIN TIME AND PROTHROMBIN TIME AS PREDICTORS OF BLEEDING IN DENGUE FEVER .

Principal Investigator:-

.

Procedure:

If you agree to be part of the research study, you will be asked the relevant history and will be subjected to relevant clinical examination and investigations. 5 ml of blood is taken for investigation. Dengue profile (NS1 & IgM) , platelet count , PT and Aptt are sent from the blood sample.

Risk and Benefits:

The only risk and possible discomfort you might get is while taking blood from your arm for the investigations. It may cause swelling, pain, redness (rarely happens) at the site from where the blood is drawn.You may not be benefitted by these investigations but you will be part of this study which is going to be useful to others in the future.

Privacy and Confidentiality:

All information collected about you during the course of this study will be kept confidential to the extent permitted by law. The code numbers will identify you in this research record. Information from this study may be published but your identity will be confidential in any publication.

I voluntarily agree to take part in this study by signing below. I may withdraw at any time. I am not giving up any of my legal rights by signing this form. My signature below indicates that I have read this consent form, or it has Been read to me, this consent form and have had all the questions answered.

Signature / Left Thumb print of the Participant or legally authorized representative

Participant's name :……...

Signature / Left thumb impression:……...

of the participant

Witness' name :……...

Signature / Left thumb impression :……...

Investigator's name and signature :……...

Date:

Place:

K.L.E.UNIVERSITY'S
JAWAHARLAL NEHRU MEDICAL COLLEGE,
NEHRU NAGAR, BELAGAVI-590010 (KARNATAKA-INDIA)
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Fax No. +91 (0)831 – 2470759

Ref: MDC/DOME/ 48

Date: 22/11/2017

To,

REG NUMBER : BG0117006

PG student in. Medicine
J.N.Medical College,
BELAGAVI.

Sub: Institutional Ethical Clearance for the study.

With reference to the above, we wish to inform you that your proposed research project titled **"STUDY OF PARTIAL THROMBOPLASTIN TIME AND PROTHROMBIN TIME AS PREDICTORS OF BLEEDING IN DENGUE FEVER – ONE EAR OF CROSS SECTIONAL STUDY"**, is ethical and justifiable. The proposed research project has been cleared by the JNMC Institutional Ethics Committee on Human Subjects Research.

(Dr. Arathi Darshan)
Member Secretary
JNMC Institutional Ethics Committee
on Human Subjects Research,
J.N.Medical College, Belagavi.

(Dr. Roopa M Bellad)
Chairman,
JNMC Institutional Ethics Committee
on Human Subjects Research,
J.N.Medical College, Belagavi.

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ASE NO	AGE	SEX	IP NO	DIAGNOSIS	INVESTIGATION																			
					NS 1/IGM	HB	PCV	TC	PLATELET	UREA	S.CREAT	T.BILIRUBIN	D.BILIRUBIN	SGOT	SGPT	ALP	T.PROTEIN	AIBUMIN	Na	K	Cl	PT (SECONDS)		APTT (SECONDS)	
						(g/dl)	(%)	per/mic	/microml	(mg/dl)	(mg/dl)	(mg/dl)	(mg/dl)	(U/L)	(U/L)	(U/L)	(gm/dl)	(gm/dl)	(meq/L)	(meq/L)	(meq/L)	C	TEST	C	T
1	29	M	899222	DF	NS1	15.4	40	1600	68000	22	1	0.49	0.19	69	53	40	6.5	4	140	3.88	102	11.2	12	28.8	33.1
2	46	F	900985	DHF	IgM	7.3	41	10000	40000	47	0.54	0.4	0.08	192	58	51	4.6	2.5	127	5.17	94	11.2	13	28.8	33
3	57	M	909167	DHF	NS1	16.5	43.8	4500	27000	26	0.99	0.2	0.06	65	38	78	7.3	4.4	135	5.69	97	11.2	11.4	28.8	38.7
4	36	F	909169	DHF	IgM	12.3	40	3100	21000	12	0.68	0.4	0.3	113	72	96	6.6	3.5	140	3.7	104	11.2	14.1	28.8	42.5
5	26	M	908893	DHF	NS1	14.3	40.2	2800	31000	17	0.96	0.3	0.09	43	53	55	6	3.8	142	4.07	104	11.2	14	28.8	36.2
6	30	M	911597	DF	IgM	13.9	41	8600	96000	20	0.9	0.5	0.1	46	33	77	6.5	3.9	135	4.2	96	11.2	11.5	28.8	30.5
7	18	M	912532	DHF WITH ALI	IgM	14.4	43.5	6500	9000	23	0.68	1.28	0.98	350	125	220	6.1	3.6	136	4.39	100	11.2	14.2	28.8	38.5
8	40	M	875518	DHF	NS1	13.7	40	6600	44000	18	1.04	1.2	0.3	27	17	50	6.4	4.1	136	3.9	98	11.2	11.3	28.8	33.3
9	25	M	919038	DHF WITH ALI	IgM	12.3	44	8800	30000	18	1.57	2.9	2.7	352	169	116	5.8	3.1	142	4.38	103	11.2	16.2	28.8	39.2
10	20	F	803875	DSS	IgM	6.4	41	22800	5000	32	0.95	1.41	0.54	90	28	67	5.6	2.8	141	3.99	104	11.2	11	28.8	38.4
11	26	M	867868	DHF	NS1	14	40	2300	28000	16	0.89	0.42	0.22	318	182	59	5.8	3.4	138	4.22	101	11.2	11.3	28.8	50.2
12	23	M	867844	DF	NS1	14.9	41.6	3700	78000	17	0.86	0.79	0.32	33	24	84	7.1	4.3	142	4.75	102	11.2	14.4	28.8	43.2
13	21	M	867852	DF	IgM	13.7	41	3200	94000	28	1.01	0.72	0.28	80	54	51	6	3.2	124	3.81	88	11.2	12	28.8	27.02
14	38	M	869024	DHF	IgM	14.6	44.6	7400	58000	13	0.63	0.61	0.14	66	75	99	7.2	4.1	138	3.59	99	11.2	11.4	28.8	33.6
15	22	F	876039	DHF	NS1	14	41	3200	45000	16	0.5	0.3	0.1	151	96	62	6.7	3.9	142	3.4	105	11.2	11.4	28.8	45.5
16	59	M	887836	DHF	IgM	13.6	44	10700	70000	42	0.67	1.4	1.1	37	43	125	6.5	3.8	132	4.2	93	11.2	12.2	28.8	32.8
17	75	F	887188	DHF	NS1	11	40	12600	92000	37	1.27	0.4	0.1	33	15	57	8.3	4.3	137	4.23	96	11.2	11.2	28.8	33
18	38	M	876943	DHF	IgM	14.8	43	3300	11000	17	0.81	0.7	0.3	144	88	133	6	3.7	140	4.01	105	11.2	12	28.8	46.5
19	25	M	888351	DHF	NS1	15.5	43.2	2600	49000	23	1.1	0.4	0.1	98	25	45	6.5	4.2	136	4.4	99	11.2	11.4	28.8	34.1
20	52	F	872057	DHF	IgM	13.2	42.7	12300	98000	10	0.33	0.25	0.11	179	148	75	6.5	3.8	136	4.56	97	11.2	10.4	28.8	42
21	18	M	881100	DHF	NS1	14.1	41	7100	53000	27	1.15	0.54	0.15	23	32	77	7.5	4.6	141	4.19	100	11.2	17.9	28.8	31.4
22	23	M	880916	DF	NS1	14.6	40	6300	95000	23	1.05	0.4	0.15	22	28	88	7	4.3	139	3.9	101	11.2	11	28.8	36.1
23	45	M	878357	DHF WITH ALI	NS1	11.2	45	4200	38000	32	1.42	4.78	3.87	319	174	119	5.1	2.9	134	3.8	96	11.2	14.9	28.8	56.9
24	18	M	825940	DHF	NS1	14.2	40	2700	71000	24	1.2	0.7	0.2	45	37	65	5.9	4.1	136	3.8	100	11.2	16.4	28.8	39.9
25	58	M	879837	DHF	NS1	16.1	43.8	6600	13000	35	1.31	0.67	0.3	95	49	78	6.1	3.6	139	4.2	101	11.2	10.6	28.8	52.5
26	28	M	880067	DHF	NS1	15.7	48.1	2600	82000	16	0.85	0.32	0.16	104	81	97	6.8	4.2	133	3.7	96	11.2	10.3	28.8	33.3
27	28	M	880827	DHF	IgM	11.8	43.6	6200	38000	12	0.82	1.35	0.72	95	55	43	7.6	4.2	136	4.16	95	11.2	10.9	28.8	43.3
28	23	M	826158	DHF	IgM	15.8	42	4500	16000	41	0.8	0.3	0.2	256	153	136	6.8	3.7	140	5.13	105	11.2	11.5	28.8	38.6
29	25	F	881301	DHF	NS1	13.8	44	5300	22000	18	0.79	0.42	0.21	126	65	142	6.5	3.3	137	3.69	100	11.2	10.7	28.8	20.8
30	40	M	884752	DF	NS1	16.1	41	2800	82000	15	1.07	0.48	0.1	45	24	58	6.6	4	141	4.55	102	11.2	11.5	28.8	34.2
31	40	M	884406	DHF	NS1	13.5	42.4	2100	61000	36	1.02	0.3	0.12	61	42	66	6.8	4.3	137	3.95	101	11.2	10.3	28.8	34.6
32	29	M	884343	DHF	IgM	17	43	1800	23000	16	1.13	1.09	0.67	111	49	100	6	3.8	138	3.79	100	11.2	11	28.8	35.5
33	35	M	887094	DHF	NS1	11.9	40.6	2900	76000	20	1.19	0.56	0.2	31	26	58	5.8	4	139	3.13	102	11.2	14.2	28.8	32
34	38	M	826959	DHF	IgM	17.6	40	18600	6000	20	0.86	2.2	1.5	150	145	160	7.3	3.9	138	4.4	98	11.2	10.9	28.8	43.4
35	38	M	804840	DHF	IgM	15.5	43.5	8000	26000	13	0.62	0.96	0.3	202	128	62	5.7	3.7	120	4.43	82	11.2	11.4	28.8	29.8
36	19	M	884250	DHF	NS1	12.5	43.1	9700	88000	20	1.13	0.7	0.3	20	12	67	6.7	4.5	136	4.56	98	11.2	17	28.8	31.2
37	25	F	885139	DHF	IgM	12.6	44	4200	13000	12	0.44	0.5	0.3	97	36	220	6	3	134	4.19	101	11.2	11.2	28.8	41.2
38	11	F	885510	DHF	NS1	11.2	41	4500	92000	22	0.7	0.5	0.2	32	15	152	7.6	4.2	133	4.1	96	11.2	12.6	28.8	38.6
39	19	F	884238	DHF	IgM	12.9	42	4700	39000	10	0.4	0.4	0.1	118	58	90	6.6	3.3	138	4.2	106	11.2	13.2	28.8	36.6
40	17	M	885244	DSS	IgM	20	42.6	6100	9000	43	0.9	0.6	0.05	53	21	172	8.1	4.6	133	6.4	92	11.2	12.8	28.8	54.2
41	22	M	828349	DHF	NS1	15.7	42	3600	16000	33	1	0.9	0.2	93	47	102	6.2	4	135	4.19	94	11.2	11.6	28.8	48.4
42	36	F	884237	DHF	IgM	16.3	44	6400	22000	28	0.7	1.4	1.1	472	226	523	6.1	2.9	132	3.2	100	11.2	10.4	28.8	48.8
43	18	M	884661	DHF	IgM	13.1	40.5	4600	64000	25	0.9	0.5	0.2	51	19	68	7.1	4.1	139	4.3	100	11.2	11.6	28.8	43
44	16	M	885827	DHF	NS1	16.1	41	2100	36000	22	0.9	0.3	0.1	211	42	115	7.1	4.4	138	4.3	100	11.2	13.2	28.8	44
45	40	M	828921	DHF WITH ALI	IgM	20.1	40	3300	5000	14	0.9	0.4	0.3	473	385	101	6.9	4.5	137	4.9	97	11.2	11.9	28.8	53.7
46	42	M	886015	DHF	IgM	12.1	48	5800	84000	11	0.9	0.6	0.2	50	55	57	7.3	4.4	148	4.5	109	11.2	11.6	28.8	38.2
47	41	F	883456	DF	NS1	12.8	43	3200	90000	22	0.8	0.3	0.1	22	25	85	7	4.1	138	4.3	100	11.2	12.3	28.8	37.5
48	29	M	885683	DHF	IgM	17.8	41	3100	13000	17	0.7	0.8	0.3	85	50	74	6.5	3.6	136	4.3	100	11.2	11.4	28.8	35
49	26	F	885865	DHF	NS1+IgM	13.2	44.6	4100	91000	18	0.7	0.4	0.1	36	17	45	7.1	4.3	134	3.7	96	11.2	12.1	28.8	37
50	20	F	885259	DHF WITH ALI	NS1	13.9	44	6200	98000	12	0.76	2.13	1.9	3287	3671	203	6.5	3.5	137	3.76	101	11.2	16.2	28.8	34.7
51	40	M	884373	DHF	IgM	16.5	41	3300	21000	37	0.98	0.66	0.33	193	151	83	6.7	4	136	3.89	97	11.2	11.2	28.8	43.2
52	53	M	886437	DF	NS1	10.8	41	10700	78000	22	0.88	0.5	0.2	18	15	73	6.6	4.1	129	3.73	93	10.2	10.3	28.8	29.6

53	59	M	887039	DHF	IgM	16	41.8	4700	18000	29	0.89	0.5	0.2	258	208	75	6.7	4	135	4.08	97	11.2	12.9	28.8	36.2
54	52	M	885861	DSS WITH ALI	NS1	18.8	42	3700	11000	54	0.9	5.19	3.42	460	158	230	6.2	3.5	132	4.89	95	11.2	12.9	28.8	49.8
55	21	M	888318	DHF	NS1	16.7	41	5600	41000	26	1.14	0.4	0.2	97	72	62	7.4	4.2	134	4.5	98	11.2	11.9	28.8	36.9
56	56	M	888472	DF	IgM	12.4	46	3700	86000	40	1.16	1.4	0.8	52	52	105	6.2	3.6	140	3.68	103	11.2	12.4	28.8	30.9
57	20	M	889886	DHF	IgM	14.7	41	2500	63000	12	0.87	0.2	0.1	123	75	170	6.3	4.4	140	3.71	104	11.2	13.2	28.8	41.2
58	24	M	890619	DHF	IgM	14	41.3	7100	30000	34	0.82	0.7	0.1	150	51	54	6.8	4.4	131	4	94	11.2	11.7	28.8	57.9
59	33	M	890434	DHF	NS1	14.2	41.6	2500	89000	21	0.97	0.3	0.1	76	32	81	6.6	4.2	138	4.2	101	11.2	15.1	28.8	42.8
60	30	F	896312	DHF	IgM	11.4	43	2800	93000	19	0.6	0.4	0.1	89	29	55	6.9	3.7	138	2.9	100	11.2	10.5	28.8	35.6
61	21	M	829948	DSS	IgM	14.9	42	2800	10000	21	0.8	0.3	0.1	71	40	75	6.7	4	136	4.1	96	11.2	19.3	28.8	58.5
62	25	M	831957	DHF	IgM	12.7	44	3100	21000	10	0.5	0.9	0.4	448	272	205	6.9	4	133	4.4	104	11.2	13.5	28.8	50.5
63	28	M	896656	DHF	NS1	12.5	41	5300	26000	25	0.94	0.5	0.2	342	224	105	7.4	4.1	139	5.08	98	11.2	10.7	28.8	34.4
64	27	F	895229	DHF	IgM	13.8	40.1	6700	16000	11	0.67	1.5	0.9	329	231	121	5.4	3.1	140	4.54	102	11.2	10.1	28.8	39.5
65	49	M	895344	DHF	IgM	15.4	41	3900	19000	21	0.97	0.6	0.2	54	38	52	6.3	3.7	144	3.2	106	11.2	10.1	28.8	38.7
66	40	M	895214	DHF	NS1	14	40	2800	58000	18	0.96	0.5	0.2	119	82	74	6.6	3.8	131	3.89	92	11.2	12	28.8	43.1
67	46	F	895230	DHF	NS1	12.9	44	2800	92000	14	0.51	0.3	0.1	36	32	60	6.8	4.2	142	3.67	105	11.2	14.4	28.8	39.7
68	18	F	895228	DHF	NS1	12.1	48.9	3700	50000	11	0.59	0.3	0.1	19	13	46	6.7	4.2	140	4.38	103	11.2	13.2	28.8	39.9
69	45	M	895336	DHF WITH ALI	NS1	12.2	44.6	13000	27000	97	1.15	5.2	4.2	50	35	93	5.2	2.8	139	4.84	103	11.2	12.7	28.8	33.4
70	40	M	895918	DHF WITH ALI	IgM	14.9	41	14900	18000	88	1.27	5.71	4.99	124	118	317	5.4	2.6	133	3.57	96	11.2	12.5	28.8	32.5
71	18	M	896006	DHF	IgM	14.5	41.5	4900	35000	17	0.79	0.5	0.2	51	28	237	6.8	3.6	141	4.61	108	11.2	12.1	28.8	36.5
72	23	F	896316	DHF	NS1	12.7	46.2	2100	32000	10	0.49	0.3	0.1	39	16	29	6	3.5	133	3.39	97	11.2	11.3	28.8	47.8
73	21	M	896768	DHF	IgM	15.7	41	6200	38000	26	1.02	1.43	0.56	291	188	80	6.1	4.1	134	3.83	85	11.2	12.2	28.8	48.2
74	32	F	897019	DF	IgM	13.8	42	6900	23000	26	0.78	0.3	0.1	146	65	57	6.3	3.2	136	5.12	102	11.2	10.4	28.8	27
75	21	M	897758	DHF	IgM	18.3	41	10200	22000	18	0.86	0.6	0.2	127	101	87	5.2	3	134	4.77	99	11.2	10.6	28.8	39.2
76	24	M	898997	DHF	IgM	14.8	43	4900	37000	24	1.03	0.4	0.1	48	29	49	6.4	3.9	139	3.56	99	11.2	11.5	28.8	31.5
77	44	M	832690	DF	NS1	15.1	40	5000	148000	17	0.8	0.5	0.2	48	34	65	7.5	4.6	137	4.2	96	11.2	11.4	28.8	27.9
78	35	M	924015	S WITH ENCEPHAL	NS1	16.5	40.5	8000	6000	27	1.02	0.5	0.3	38	40	94	8.6	5.1	131	4.07	100	11.2	11.4	28.8	44.7
79	25	M	838549	DHF	NS1	18.3	42	4300	72000	35	1.2	1.08	0.5	106	113	147	6.5	3.9	130	3.2	90	11.2	11.2	28.8	47.2
80	19	M	878673	DF	IgM	15.6	45	7100	98000	18	0.92	0.4	0.1	67	52	63	7.1	4.2	132	3.5	97	11.2	14.4	28.8	33.8
81	30	F	879947	DHF	IgM	12.1	41	3800	35000	12	0.4	0.4	0.2	87	68	128	7.5	4.2	132	4.2	99	11.2	11.7	28.8	35.1
82	21	M	884220	DHF	NS1	14.7	42	8200	74000	21	0.94	0.9	0.2	80	28	126	7.5	4.9	140	4.68	97	11.2	12.5	28.8	40.9
83	21	F	834503	DHF	NS1	14.8	38	3700	30000	23	0.6	0.4	0.2	41	79	56	5.9	3.4	137	3.8	101	11.8	12.5	28.8	45.2
84	34	M	892462	DHF	IgM	14.2	41	9100	13000	28	1.11	1.29	0.74	179	106	77	6.6	3.6	137	4.48	99	11.2	11.6	28.8	45.8
85	38	M	895397	DHF WITH ALI	IgM	13.6	38.6	12600	16000	38	0.38	3.25	1.96	9065	2839	205	5.3	3	135	5.23	98	11.2	19.7	28.8	50.2
86	30	M	839818	DHF	IgM	15.6	40	6000	2000	18	0.9	0.4	0.1	35	31	52	7.3	3.8	138	4.4	101	11.2	12.1	28.8	34.3
87	24	M	841152	DHF	NS1	15.8	37	4800	136000	13	1.02	0.5	0.3	93	110	110	6.8	4.2	135	3.9	95	11.2	12.5	28.8	48.8
88	28	F	841922	DHF	IgM	13.7	41.7	11300	18000	13	1.02	0.4	0.2	130	121	59	6.6	3.7	138	4.4	100	11.2	11.4	28.8	48.7
89	20	M	844910	DHF	NS1	14.6	43.1	4400	14000	16	0.7	0.6	0.3	192	128	79	6.4	3.3	134	4.08	93	11.2	105	28.8	39.8
90	24	M	845070	DF	NS1	13.3	40	3500	80000	25	1.34	0.4	0.1	30	16	74	6.8	4.1	134	3.8	95	11.2	10.9	28.8	34.7
91	31	M	910652	DHF	IgM	16.8	41	6300	18000	27	1.02	1.12	0.6	75	57	142	6.2	3.8	134	3.51	94	11.2	11.4	28.8	38.7
92	26	F	912517	DHF	IgM	10.2	41	5400	61000	13	0.67	0.4	0.2	61	50	147	6.9	3.3	139	3.78	101	11.2	12.8	28.8	36.9
93	29	M	912518	DHF	IgM	17.9	39	7100	11000	13	0.9	2.34	1.97	182	124	406	6.1	3.5	136	4.19	100	11.2	11.2	28.8	41
94	54	M	846862	DF	NS1	14.1	44	3800	131000	25	1.1	0.4	0.1	21	33	90	7	4.2	135	4.08	100	11.2	12.5	28.8	36.5
95	60	F	854314	DHF	IgM	12.8	40.2	3800	102000	17	0.7	0.7	0.3	71	57	186	6.9	3.5	134	3.9	93	11.2	10.2	28.8	35.9
96	21	M	861255	DF	IgM	14.1	40.1	4700	149000	21	0.8	7.3	5.6	828	1552	170	6.8	3.9	146	4.2	104	11.2	15.2	28.8	32.1
97	29	M	872985	DHF	NS1	12.1	40.1	5700	59000	10	0.5	0.4	0.1	50	28	53	6.6	3.7	136	4.3	99	11.2	11.4	28.8	39.2
98	45	M	870082	DHF	IgM	14.5	40	3000	6000	13	1.02	1.09	0.65	273	193	184	5.5	3.2	136	4.7	98	11.2	10.8	28.8	34.9
99	24	F	911096	DHF	IgM+NS1	15	44.6	5400	12000	21	0.67	0.48	0.1	78	49	59	7.1	3.9	137	4.34	98	11.2	10.8	28.8	37.9
100	30	M	910252	DF	NS1	15	46.2	2400	140000	14	1.44	0.4	0.2	45	16	72	7.4	4.5	136	3.71	95	11.2	11.2	28.8	34.5