

**ORIGINAL RESEARCH****Correlation of D-dimer levels with severity of dengue fever**

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**ABSTRACT**

**Introduction:** Thrombocytopenia and bleeding are common complications of dengue fever. Platelet count is a poor predictor of bleeding manifestations. A surrogate marker of bleeding can be D-Dimer assay which is often used to describe coagulation activity in a patient. This project aims to study the correlation of D-dimer level with coagulation parameters so that d-dimer assay can be considered as an important predictor and prognostic marker for development of hemorrhage or shock in dengue.

**Methodology:** This was a cross sectional study done on dengue patients attending Gandhi hospital from 2019 to 2021. Patients were grouped in to 2 groups – group 1 having no bleeding manifestations and group 2 having bleeding manifestations. D-dimer levels in both groups were compared with platelet count, aPTT, PT and INR. Means of both groups were compared using student's unpaired t-test. p value of <0.05 was taken as significant. Correlation of d-dimer with thrombocytopenia and coagulation parameters was done using Pearson's correlation

**Results:** 100 patients were recruited in this study. Of these 100, 57 patients did not have any bleeding manifestation and 43 had bleeding manifestations. Mean age in group 1 was 40.60±16.14 and in group 2 was 38.95±15.60. Group 1 had 23 (40.35%) females and group 2 had 17 (39.53%) females. Mean platelet count in group 1 was 72,263.16±26,340.85 /ul while in group 2 was 54,813.95±16,844.18 /ul. Mean APTT in group 1 was 36.65±2.02 seconds while in group 2 was 37.67±2.22 seconds. Mean APTT in group 1 was 36.65±2.02 seconds while in group 2 was 37.67±2.22 seconds. Mean PT in group 1 was 15.46±1.49 seconds while in group 2 was 16.16±1.79 seconds. Mean INR in group 1 was 1.41±0.14 while in group 2 was 1.80±0.20. Mean D-Dimer in group 1 was 929.74±268.46 ng/ml while in group 2 was 5034.53±2024.85 ng/ml. All parameters were significantly higher in group with bleeding compared to non-bleeding (p>0.05). D dimer had positive correlation with thrombocytopenia, APTT, PT and INR. Association of d dimer with APTT, PT and INR was higher in bleeding group.

**Conclusion:** D-dimer levels were raised in dengue patients with bleeding. Strong association of d-dimer levels with thrombocytopenia, APTT, PT and INR was seen in both groups. Hence it can be concluded that detection of D dimer in febrile stage of dengue infection may be beneficial for predicting the clinical course of the disease.

**Keywords:** D dimer, Activated Plasma Thromboplastin Time, thrombocytopenia, Dengue infection

## INTRODUCTION

Dengue is the most rapidly spreading mosquito-borne viral disease, with a 30-times increase in global incidence over the last five decades. Dengue is a major public health problem throughout the tropical and subtropical regions of the world. Half the world's population almost lives in countries where dengue is endemic. According to WHO, about 50–100 million new infections are estimated to occur annually in more than hundred endemic countries, with an intense increase in the number of countries reporting the disease. About 2.5% of those affected die. During epidemic, infection rate among those who have not been previously encountered to the virus are often 40 to 50 % but can also reach 80 to 90 percent. *Aedes aegypti* is the primary epidemic vector.<sup>1</sup>

Generally, Dengue is an acute febrile illness, and sometimes biphasic fever with severe myalgia, headache, arthralgia, rashes, thrombocytopenia and leucopenia may also be observed. Dengue haemorrhagic fever is characterized by acute onset of high fever and with associated signs and symptoms similar to dengue fever in the early febrile phase. There are common haemorrhagic manifestations such as positive tourniquet test (TT), petechiae, easy bruising and/or GI haemorrhage in severe cases. By the end of the pyrexia phase, there is a tendency to develop hypotensive shock (DSS) due to plasma leakage. The presence of preceding warning signs such as lethargy or restlessness or irritability, persistent vomiting, oliguria and abdominal pain are important for intervention to prevent shock. Plasma leakage and abnormal haemostasis are the main pathophysiological hallmarks of Dengue haemorrhagic fever. Rising haematocrit/ haemoconcentration and Thrombocytopenia are constant findings before the cessation of fever/ onset of shock. We live in a country with resource limited settings, added on by public ignorance about the disease and poor access to health care.<sup>2,3</sup>

The demographic pattern and the trend of dengue are largely changing every year through the past decade. South India had witnessed several dengue fever epidemic outbreaks during the past few years. Thrombocytopenia and bleeding are common complications of dengue fever. In general, platelet count is a poor predictor of bleeding manifestations. Thrombocytopenia can be due to antibody mediated platelet destruction, peripheral sequestration or transient bone marrow suppression. Haemorrhage may be a consequence of the thrombocytopenia and associated platelet dysfunction or disseminated intravascular coagulation. In addition, the bleeding severity has not been well studied in India, especially in relation to coagulation and fibrinolytic pathways. There is a dearth of laboratory parameters to predict bleeding episodes. A common surrogate marker of bleeding can be D-Dimer assay which is often used to describe coagulation activity in a patient. This parameter has not previously been assessed in dengue fever. Dengue shock syndrome and dengue hemorrhagic fever are serious complications. Additional studies about dengue can lead to change in guidelines and alterations in public health programs. Early recognition and prompt initiation of appropriate management is vital. This project aims to study the correlation of D-dimer level with coagulation parameters so that d-dimer assay can be considered as an important predictor and prognostic marker for development of hemorrhage or shock in dengue.

## PATIENTS AND METHODS

It is a Cross Sectional study was conducted from 2019 to 2021 in Department of medicine, Gandhi hospital, Secunderabad. 100 dengue patients admitted in Department of medicine, Gandhi hospital.

## INCLUSION CRITERIA

All dengue serology positive patients with thrombocytopenia (platelets < 1.5lakh) with bleeding manifestations and without bleeding manifestations.

## EXCLUSION CRITERIA

Dengue serology negative patients, Chronic kidney or liver disease,, Autoimmune or chronic infectious disease,Haematological disorders and neoplasia, Pregnant women **and** Diagnosed cases of DVT, PTE, CSVT.

Ethical clearance was obtained from IEC, Gandhi medical college and Gandhi hospital, secunderabad. Admitted dengue patients positive for NS1 Antigen or Dengue IgM antibody were enrolled in study after explaining the study procedure to the patients and obtaining informed consent from them. Detailed history taking and examination were done. Patients were grouped in to 2 groups – group1 having no bleeding manifestations and group 2 having bleeding manifestations. D-dimer levels in both groups were compared with platelet count, aPTT, PT and INR. **All basic investigations are done**

## STATISTICAL ANALYSIS

All data was collected and recorded in MS excel sheet. Data was expressed as descriptive statistics. Mean  $\pm$  SD for age, platelet count, aPTT, PT, INR and D-dimer were calculated. Statistical analysis was done using statistical commands of MS excel. Means of both groups were compared using student's unpaired t-test. p value of  $<0.05$  was taken as significant. Correlation of d-dimer with thrombocytopenia and coagulation parameters was done using Pearson's correlation where  $R^2$  of  $>0.5$  was taken as positive correlation and  $<0.5$  was taken as negative correlation.

## RESULTS

100 patients were recruited in this study after carefully applying inclusion and exclusion criteria. Of these 100, 57 patients did not have any bleeding manifestation which were grouped as group 1 and 43 had bleeding manifestations which were grouped as group 2.

**Table-1: Demographic details in present study**

	<b>Group1 : without bleeding manifestations</b>	<b>Group-2 : with bleeding manifestations</b>	
			<b>P value</b>
<b>AGE (Y)</b>	40.60 $\pm$ 16.14	38.95 $\pm$ 15.60	0.61
<b>Gender (M:F)</b>	34 (59.65%):23(40.35%)	26 (60.47%):17(39.53%)	0.93

Mean age (years) in group 1 was 40.60 $\pm$ 16.14 while that in group 2 was 38.95 $\pm$ 15.60. There was no statistically significant difference between these 2 groups based on age. With regards to gender distribution, group 1 had 34 (59.65%) males and 23 (40.35%) females. While group 2 had 26 (60.47%) males and 17 (39.53%) females.

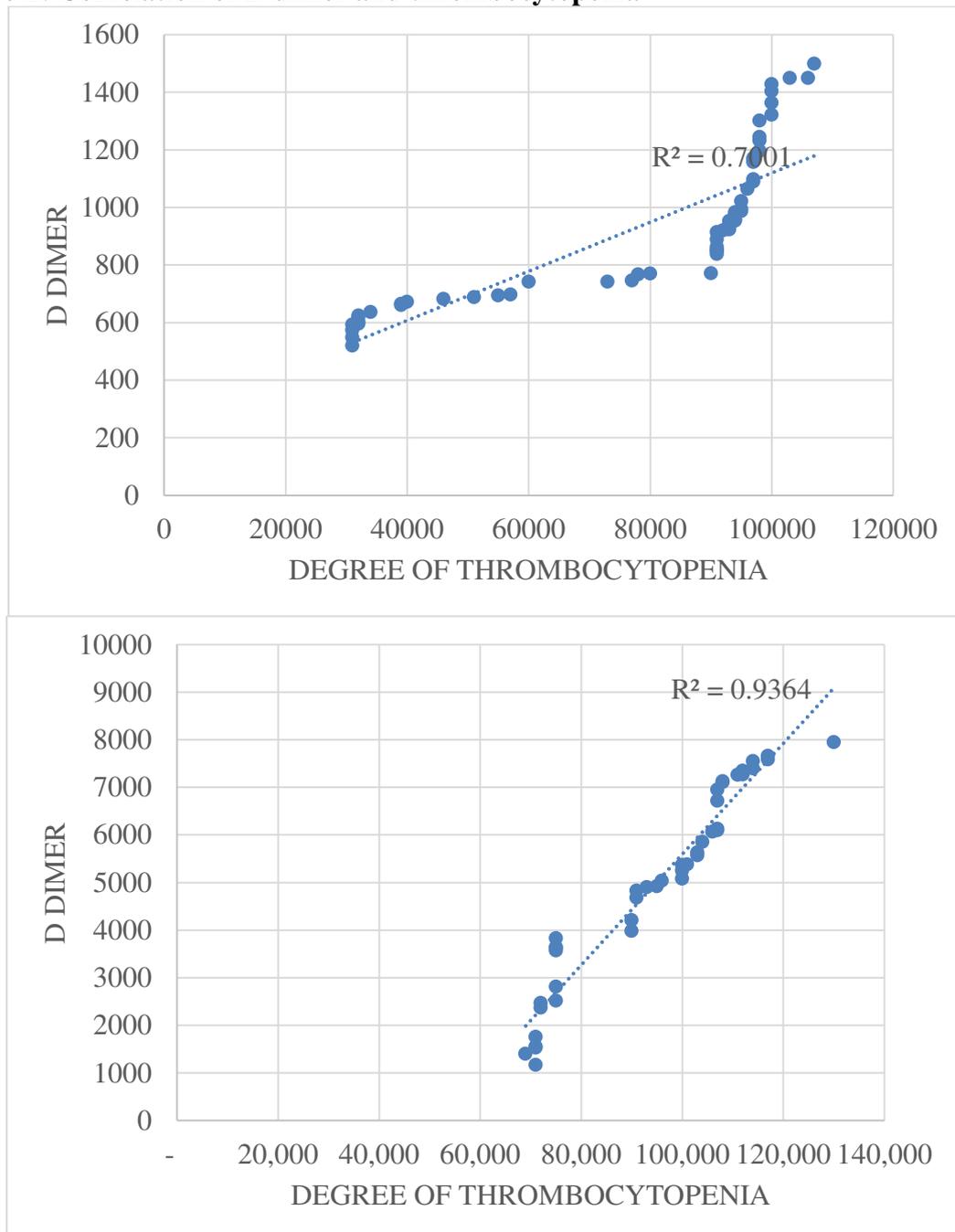
**Table 2: Coagulation Profile**

	<b>Group1 : without bleeding manifestations</b>	<b>Group-2 : with bleeding manifestations</b>	<b>P value</b>
<b>Platelet Count (cells/microL)</b>	72,263.16 $\pm$ 26,340.85	54,813.95 $\pm$ 16,844.18	<b>&lt;0.005</b>
<b>APTT (s)</b>	36.65 $\pm$ 2.02	37.67 $\pm$ 2.22	<b>&lt;0.005</b>
<b>PT (s)</b>	15.46 $\pm$ 1.49	16.16 $\pm$ 1.79	<b>&lt;0.005</b>
<b>INR</b>	1.41 $\pm$ 0.14	1.80 $\pm$ 0.20	<b>&lt;0.005</b>
<b>D-Dimer (ng/ml)</b>	929.74 $\pm$ 268.46	5034.53 $\pm$ 2024.85	<b>&lt;0.005</b>

Mean platelet count in group 1 was 72,263.16 $\pm$ 26,340.85 /ul while in group 2 was 54,813.95 $\pm$ 16,844.18 /ul. There was statistically significant difference between these 2 groups with regards to platelet count based on unpaired student's t-test. (p $<$ 0.05). Mean APTT in group 1 was 36.65 $\pm$ 2.02 seconds while in group 2 was 37.67 $\pm$ 2.22 seconds. There was statistically significant difference between these 2 groups with regards to APTT based on unpaired student's t-test. (p $<$ 0.05). Mean PT in group 1 was 15.46 $\pm$ 1.49 seconds while in

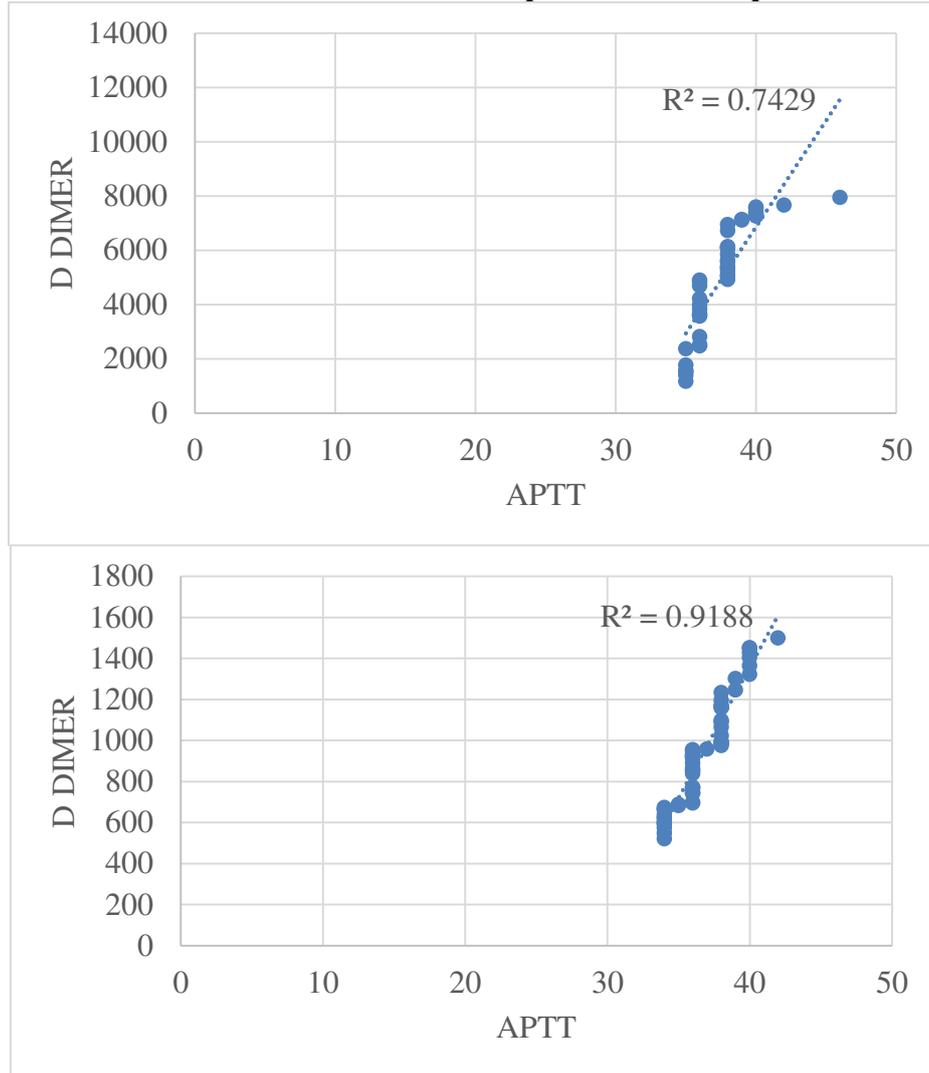
group 2 was  $16.16 \pm 1.79$  seconds. There was statistically significant difference between these 2 groups with regards to PT based on unpaired student's t-test. ( $p < 0.05$ ). Mean INR in group 1 was  $1.41 \pm 0.14$  while in group 2 was  $1.80 \pm 0.20$ . There was statistically significant difference between these 2 groups with regards to INR based on student's unpaired t-test. ( $p < 0.05$ ). Mean D-Dimer in group 1 was  $929.74 \pm 268.46$  ng/ml while in group 2 was  $5034.53 \pm 2024.85$  ng/ml. D-dimer levels were significantly raised in patients with bleeding manifestations compared to those without bleeding manifestations based on student's unpaired t-test. ( $p < 0.05$ ).

**Figure-1: Correlation of D dimer and thrombocytopenia**



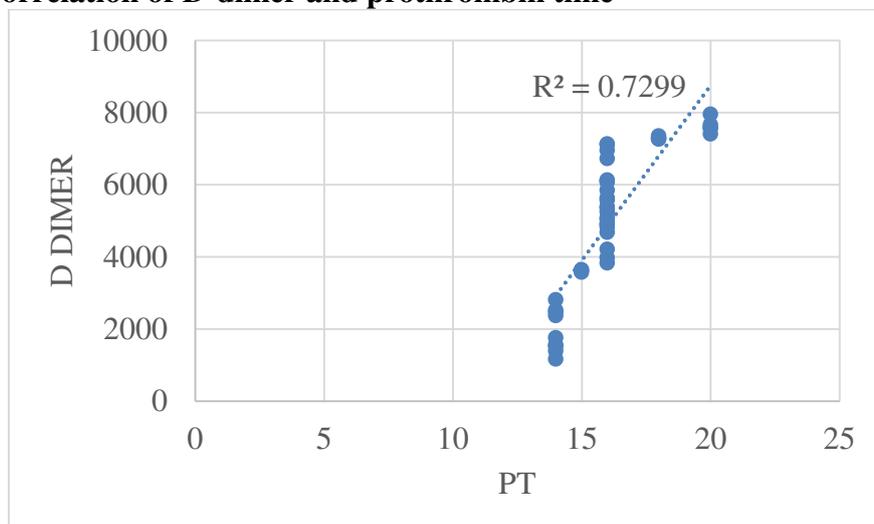
Correlation between D Dimer levels and thrombocytopenia was assessed using Pearson's correlation and it was found that d dimer had positive correlation with thrombocytopenia. R value of 0.82 and 0.96 was obtained for group 1 and group 2 respectively.

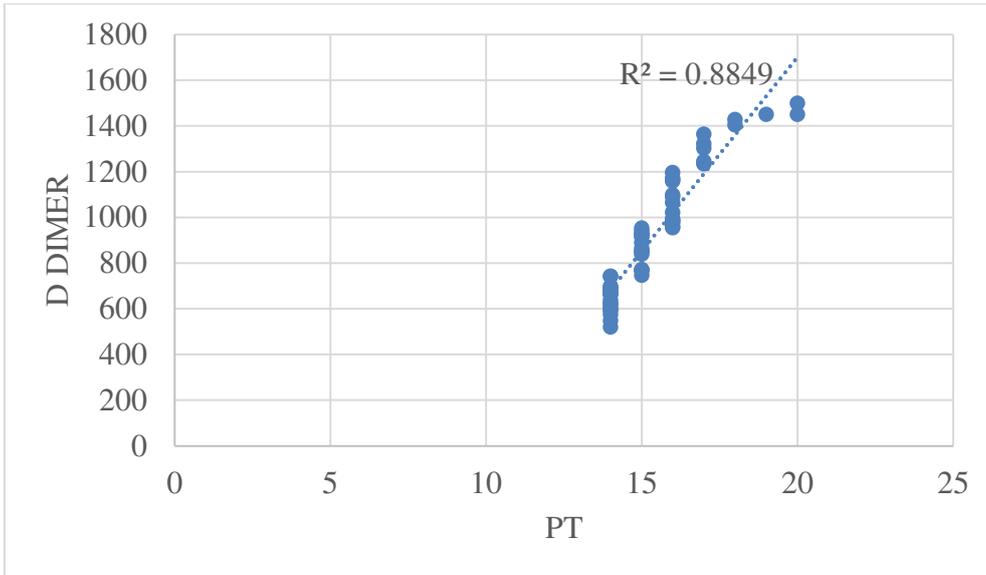
**Figure-2: Correlation of D dimer and activated plasma thromboplastin time**



Correlation between D Dimer levels and APTT was assessed using Pearson’s correlation and it was found that d dimer had positive correlation with APTT. R value of 0.86 and 0.95 was obtained for group 1 and group 2 respectively.

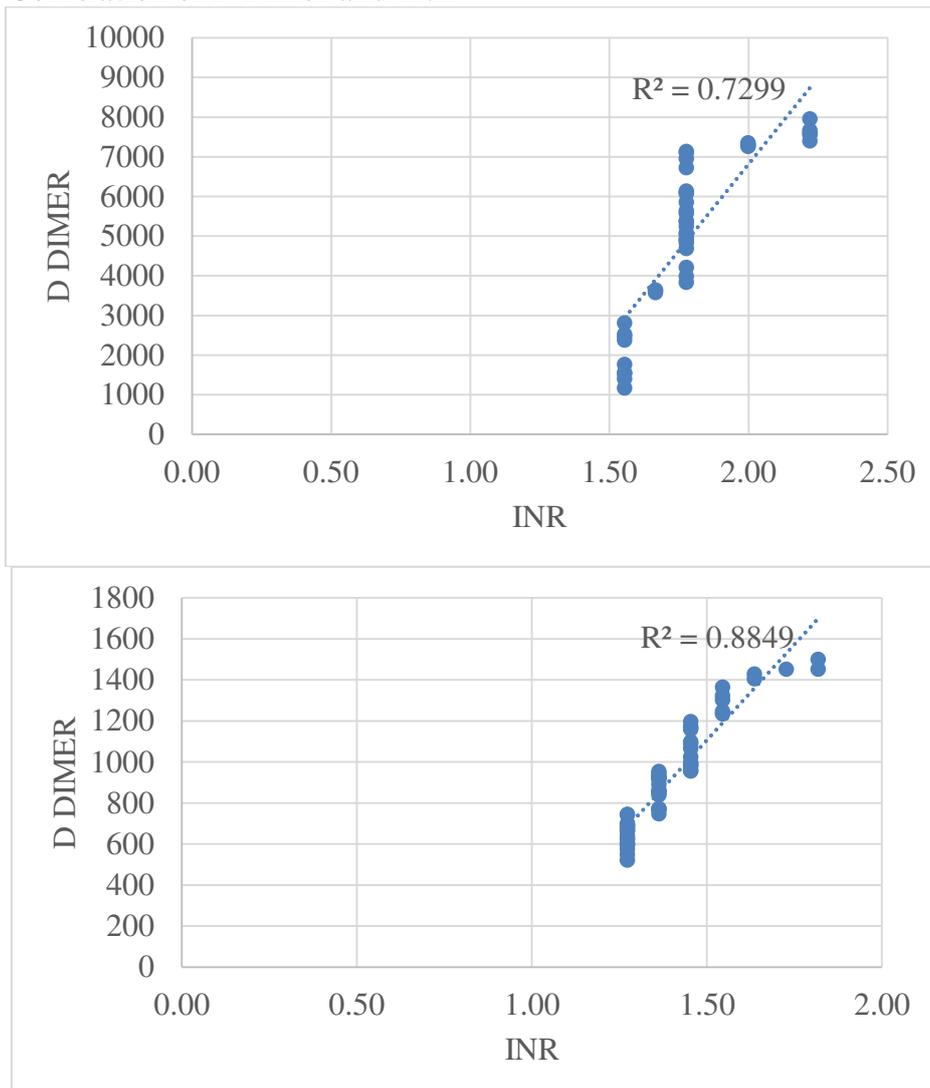
**Figure-3: Correlation of D-dimer and prothrombin time**





Correlation between D Dimer levels and PT was assessed using Pearson’s correlation and it was found that d dimer had positive correlation with PT. R value of 0.85 and 0.94 was obtained for group 1 and group 2 respectively.

**Figure-4: Correlation of D Dimer and INR**



Correlation between D Dimer levels and INR was assessed using Pearson's correlation and it was found that d dimer had positive correlation with INR. R value of 0.85 and 0.94 was obtained for group 1 and group 2 respectively.

**Table 3: Association of D dimer with coagulation parameters**

	<b>Group1 : without bleeding manifestations</b>	<b>Group-2 : with bleeding manifestations</b>
<b>Platelet Count</b>	83.66%	96.76%
<b>APTT</b>	86.19%	95.85%
<b>PT</b>	85.43%	94.06%
<b>INR</b>	85.43%	94.06%

## DISCUSSION

This study was done to study the prognostic use of D-dimer in dengue fever by correlating d dimer levels with coagulation parameters – thrombocytopenia, APTT, PT and INR. Study was done in patients not having bleeding manifestations (group 1) and in patients having bleeding manifestations (group 2).

Mean age (years) in group 1 was  $40.60 \pm 16.14$  while that in group 2 was  $38.95 \pm 15.60$ . There was no statistically significant difference between these 2 groups based on age. This shows that age is not a factor for developing bleeding manifestation in dengue. Patient of any age group can develop either hemorrhagic or non-hemorrhagic dengue. This has been reported by Tantawichien.<sup>4</sup> Similarly sex ratio was similar in both groups. Group 1 had 34 (59.65%) males and 23 (40.35%) females. While group 2 had 26 (60.47%) males and 17 (39.53%) females. Hence sex cannot be taken as predisposing factor for development of hemorrhage in dengue. This point has been supported previously by Nguyen et. al.<sup>5</sup>

Mean platelet count in group 1 was  $72,263.16 \pm 26,340.85$  /ul while in group 2 was  $54,813.95 \pm 16,844.18$  /ul. Platelet count was lower than normal range in both groups but group with bleeding manifestations had much lower platelet count compared to non-bleeding group. There was statistically significant difference between these 2 groups with regards to platelet count based on unpaired student's t-test. ( $p < 0.05$ ). Mehboob Riffat et al, Azeredo et al, and Ojha et al have established the positive correlation of platelet count with hemorrhagic manifestation.<sup>6,7</sup> Vijayaraghavan et al studied 203 cases of dengue. They reported thrombocytopenia in 56.4% of DF and in 55.5% of DSS cases.<sup>8</sup>

Mean APTT in group 1 was  $36.65 \pm 2.02$  seconds while in group 2 was  $37.67 \pm 2.22$  seconds. Higher activated plasma thromboplastin time represents delay in blood coagulation which might result in bleeding. Dengue cases in both groups had longer APTT and those with bleeding manifestations had longer APTT compared to those without bleeding manifestations. There was statistically significant difference between these 2 groups with regards to APTT based on student's unpaired t-test. ( $p < 0.05$ ). Mean PT in group 1 was  $15.46 \pm 1.49$  seconds while in group 2 was  $16.16 \pm 1.79$  seconds. Dengue cases in both groups had longer Prothrombin time and those with bleeding manifestations had longer PT compared to those without bleeding manifestations. There was statistically significant difference between these 2 groups with regards to PT based on student's unpaired t-test. ( $p < 0.05$ ). Mean INR in group 1 was  $1.41 \pm 0.14$  while in group 2 was  $1.80 \pm 0.20$ . Dengue cases in both groups had higher INR and those with bleeding manifestations had higher INR compared to those without bleeding manifestations. There was statistically significant difference between these 2 groups with regards to INR based on student's unpaired t-test. ( $p < 0.05$ ).

Many previous studies have reported raised levels of these coagulation profile parameters. Kulasinghe et al reported raised APTT and INR in 83 DHF patients, 75 DF patients of which 12 DHF patients and 1 DF patient had bleeding manifestations.<sup>9</sup> Vijayaraghavan et. al. studied 203 cases of dengue. They reported prolonged APTT in 52.2% of DF and in 68.4% of DSS

cases.<sup>8</sup>Laoprasopwattana et al studied 238 dengue cases. They reported that patients of DHF had thrombocytopenia and high INR.<sup>10</sup>Adane et al conducted a meta-analysis on 42 studies with total 12,221 dengue patients to study magnitude of prolonged APTT, PT, and thrombocytopenia. They reported that 70.29% dengue cases had thrombocytopenia, 42.91% dengue patients had prolonged APTT and 16.48% dengue cases had prolonged PT.<sup>11</sup>

Mean D-Dimer in group 1 was  $929.74 \pm 268.46$  ng/ml while in group 2 was  $5034.53 \pm 2024.85$  ng/ml. D-dimer levels were significantly raised in patients with bleeding manifestations compared to those without bleeding manifestations based on student's unpaired t-test ( $p < 0.05$ ). Sridhar et al had studied 60 DHF sero-positive cases. They reported that cases presenting with clinical features of shock and thrombocytopenia had significantly higher D-Dimer levels.<sup>12</sup>Orsi et al conducted a case control study on 33 dengue patients. They reported that D-dimer was significantly increased in patients with bleedings (median D-dimer levels were 515.5, 1028 and 1927 ng/ml,  $P < 0.0001$ ).<sup>13</sup>SetrkraisingKittiya et al reported that d-dimer levels correlated with severity of dengue fever and suggested that d-dimer levels could be used as indicator of dengue severity.<sup>14</sup>

To check for correlation between d-dimer and thrombocytopenia, platelet count of each patient was subtracted from lower limit of normal range of platelet count i.e. 1,50,000 to get the 'degree of thrombocytopenia' in each case. This degree of thrombocytopenia was correlated with d-dimer using Pearson's correlation and their linear regression was also plotted. It was found that d dimer had positive correlation with thrombocytopenia. R value of 0.82 and 0.96 was obtained for group 1 and group 2 respectively. Hence stronger association was observed in dengue with bleeding manifestations. Sridhar et al also reported positive correlation of d dimer with thrombocytopenia.<sup>12</sup>

Correlation between D Dimer levels and APTT, PT and INR was assessed using Pearson's correlation and it was found that d dimer had positive correlation with APTT, PT and INR. R value of 0.86 and 0.95 was obtained for group 1 and group 2 respectively for APTT. R value of 0.85 and 0.94 was obtained for group 1 and group 2 respectively for PT. R value of 0.85 and 0.94 was obtained for group 1 and group 2 respectively for INR. Hence stronger association of d dimer with APTT, PT and INR was observed in dengue with bleeding manifestations. Setrkraising et al reported positive correlation of dimer with PT, APTT and INR in dengue patients.<sup>14</sup> Kannan et al reported positive correlation of d dimer with APTT in dengue cases.<sup>15</sup>

Based on observations from this study, it can be concluded that d-dimer levels play a crucial role in predicting further course of disease in dengue fever. Which will be of great help when patient needs to be shifted to a critical care unit or higher centre if hemorrhage is expected. However our study was done only in adults at a tertiary care centre which caters to all complicated cases in our state and nearby districts of our state so these findings may be applicable only to region around us for adults. Further multi centric studies in all age groups are required to establish accurate predictability of dengue patients based on d-dimer assay.

## CONCLUSION

In this study, thrombocytopenia was more pronounced in patients of dengue fever with bleeding manifestations in comparison to patients of dengue without bleeding manifestations. This was also reflected by increased APTT, PT and INR levels in dengue patients with bleeding manifestations compared to dengue patients without bleeding manifestations. D-dimer levels were raised in dengue patients with bleeding manifestations but were in normal to near normal range in dengue patients without bleeding manifestations.

Strong association of d-dimer levels with thrombocytopenia, APTT, PT and INR was seen in both groups. This association was more prominent in dengue patients with bleeding manifestations. Hence it can be concluded that detection of D dimer in febrile stage of dengue

infection may be beneficial for predicting the clinical course of the disease. D-dimer levels may help us in predicting dengue severity before the patients progress into toxic stage so that close monitoring and proper management can be arranged.

## REFERENCES

1. Medagama A, Dalugama C, Meiyalakan G, Lakmali D. Risk Factors Associated with Fatal Dengue Hemorrhagic Fever in Adults: A Case Control Study. *Can J Infect Dis Med Microbiol.* 2020 May 5;2020:e1042976.
2. HentzyMoraes G, de Fátima Duarte E, Carmen Duarte E. Determinants of Mortality from Severe Dengue in Brazil: A Population-Based Case-Control Study. *Am J Trop Med Hyg.* 2013 Apr 3;88(4):670–6.
3. Heilman JM, Wolff JD, Beards GM, Basden BJ. Dengue fever: a Wikipedia clinical review. *Open Med.* 2014 Oct 2;8(4):e105–15.
4. Tantawichien T. Dengue Fever And Dengue Hemorrhagic Fever In Adults. *Southeast Asian J Trop Med Public Health.* 2015;46 Suppl 1:79–98.
5. Nguyen TH, Nguyen TL, Lei H-Y, Lin Y-S, Le BL, Huang K-J, et al. Association between sex, nutritional status, severity of dengue hemorrhagic fever, and immune status in infants with dengue hemorrhagic fever. *Am J Trop Med Hyg.* 2005 Apr;72(4):370–4.
6. Mehboob Riffat, Munir Muhammad, Azeem Ahmed, Naeem Samina, Ahmad FridoonJawad. LOW PLATELET COUNT ASSOCIATED WITH DENGUE HEMORRHAGIC FEVER. *Int J Adv Chem.* 2015 Nov 1;1(1):31–6.
7. Ojha A, Nandi D, Batra H, Singhal R, Annarapu GK, Bhattacharyya S, et al. Platelet activation determines the severity of thrombocytopenia in dengue infection. *Sci Rep.* 2017 Jan 31;7(1):41697.
8. Vijayaraghavan, Thay Wee Y, FoongShing W, Hafsa P. Predictors of Dengue Shock Syndrome: APTT Elevation as a Risk Factor in Children with Dengue Fever. *J Infect Dis Epidemiol [Internet].* 2020 Feb 17 [cited 2021 Dec 4];6(1).
9. Kulasinghe S, Ediriweera R, Kumara P. Association of abnormal coagulation tests with Dengue virus infection and their significance as early predictors of fluid leakage and bleeding. *Sri Lanka J Child Health.* 2016 Sep 5;45(3):184.
10. Laoprasopwattana K, Binsai J, Pruekprasert P, Geater A. Prothrombin Time Prolongation was the Most Important Indicator of Severe Bleeding in Children with Severe Dengue Viral Infection. *J Trop Pediatr.* 2017 Aug 1;63(4):314–20.
11. Adane T, Getawa S. Coagulation abnormalities in Dengue fever infection: A systematic review and meta-analysis. *PLoS Negl Trop Dis.* 2021 Aug 18;15(8):e0009666.
12. Sridhar A, Sunil Kumar BM, Rau A, Rau ATK. A Correlation of the Platelet Count with D-Dimer Levels as an Indicator for Component Therapy in Children with Dengue Hemorrhagic Fever. *Indian J Hematol Blood Transfus.* 2017 Jun;33(2):222–7.
13. Orsi FA, Angerami RN, Mazetto BM, Quaino SK, Santiago-Bassora F, Castro V, et al. Reduced thrombin formation and excessive fibrinolysis are associated with bleeding complications in patients with dengue fever: a case–control study comparing dengue fever patients with and without bleeding manifestations. *BMC Infect Dis.* 2013 Jul 28;13(1):350.
14. SetrkraisingKittiya, Bongsebandhu-phubhakdiChansuda, VoraphaniNipasiri, PancharoenChitsanu, ThisyakornUsa, ThisyakornChule. D-dimer as an indicator of dengue severity. *Asian Biomed.* 2007 Jun 1;1(1):53–7.
15. Kannan A, Narayanan K, Sasikumar S, Philipose J, Surendran S. Coagulopathy in dengue fever patients. *Int J Res Med Sci.* 2014;2(3):1070.