# Association Between Fasting Plasma Glucose and Routine Coagulation Tests

## Rutin Koagülasyon Testleri ve Açlık Plazma Glukozu Arasındaki İlişki

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**Abstract**

***Objective:*** **The main goal of this study was to determine relationship between activated partial thromboplastin time (APTT), prothrombin time (PT), fibrinogen levels and fasting plasma glucose levels (FPG).**

***Methods:*** **In 5602 patients APTT, PT, fibrinogen and FPG levels were measured by routine methods. Patients were divided into three groups according to their FPG levels (euglycemic, impaired fasting glucose group (IFG) and diabetic group).**

***Results:*** **We found significantly shorter APTT values and increased fibrinogen levels in both diabetic group and IFG group when compared with the euglycemic group.**

***Conclusion:*** **Shortened APTT and increased fibrinogen levels in both diabetic group and IFG group could be a useful marker for evaluation of hemostatic state.**

**Key Words:** Fasting plasma glucose, shortened APTT, prothrombin time

**Özet**

***Amaç:*** **Bu çalışmanın temel amacı aktive parsiyel tromboplastin zamanı (APTZ), protrombin zamanı (PZ), fibrinojen seviyeleri ve açlık plazma glukoz (APG) seviyelerinin arasındaki ilişkiyi belirlemektir.**

***Gereç ve Yöntem:*** **5602 hastada APTZ, PZ, fibrinojen ve APG seviyeleri rutin metodlar ile ölçülmüştür. Hastalar, APG seviyelerine göre üç gruba ayrılmıştır (öglisemik, bozulmuş açlık glukozu (BAG) ve diabetik grup).**

***Sonuçlar:*** **Çalışmamızda, öglisemik gruba kıyasla hem diabetic hem de BAG gruplarında önemli kısalmış APTZ ve önemli artmış fibrinojen seviyeleri bulduk.**

***Sonuç:*** **Kısalmış APTZ ve artmış fibrinojen seviyeleri hem diabetik hem de BAG grubunda hemostatik durumun değerlendirilmesinde yararlı bir belirteç olarak kullanılabilir.**

**Anahtar Kelimeler:** Açlık plazma glukozu, kısalmış APTZ, protrombin zamanı

The activated partial thromboplastin time (APTT) was first developed in 1953 (1), and further redefined in 1961, in which used kaolin as the 'contact activator' and enabled standardization and widespread implementation as a global screening assay (2). The term 'partial', reflects the lack of tissue factor from the reaction mixture. Although previously the attention has been focused on abnormal prolongations, and their potential associations with in the intrinsic pathway (3), there is increasing evidence that shortened APTTs may be detected with a relative high frequency in the hemostasis laboratory (e.g., 6–9% of routine APTTs, in laboratory which used very sensitive reagents, up to 15%) (3, 4). However

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shortened APTT has been defined as an independent risk factor for thromboembolic complications, after adjusting for sex, age, and factor VIII levels (5, 6).

Hyperglycemia related abnormalities in hemostasis would predispose to or trigger thromboembolic complications in diabetes (7). Hyperglycemia affects multiple steps of coagulation such as thrombus formation and inhibition, fibrinolysis, platelet, and endothelial function via non enzymatic glycation, increased oxidative stress and decreased heparan sulphate (8). Fibrinogen levels influence thrombogenesis, blood viscosity and platelet aggregation. Some studies have found a significant relationship between fibrinogen levels and insulin levels (9, 10). Moreover, chronic hyperglycemia and tissue glycation may effect the fibrin structure, clot generation and resistance to fibrinolysis (9). Recently, it has been reported that shortened APTT and increased fibrinogen levels might be useful hemostatic markers in diabetic patients, especially in those at high-risk for thrombotic complications (11).

In the present study, we aimed to evaluate the differences of APTT, prothrombin time (PT) and fibrinogen levels among groups based on fasting plasma glucose (FPG) and to determine whether differences are related FPG levels.

## Materials and Methods

Our study included 5602 patients who were admitted to various clinical departments in the outpatient clinics of Haseki Education and Training hospital. The all patients underwent APTT, PT, fibrinogen and FPG measurements. We retrospectively evaluated the combined levels of APTT, PT, fibrinogen and FPG which have been performed in our laboratory for routine blood testing. None of patients had any medication of anticoagulant therapy. In our study, we excluded PT and APTT values more than reference range. The groups based on FPG values as follows: euglycemic group (<5.6 mmol/L), impaired fasting glucose group (IFG, 5.6 to 6.9 mmol/L), and diabetic group (≥7 mmol/L).

APTT, PT and fibrinogen were assayed on the MDA II (Trinity Biotech, PLC Bray.Co Wicklow, Ireland) employing proprietary Trinity Biotech reagents (Platelin LS for APTT, TriniCLOT PT HTF for PT with an International Sensitivity Index of 1.22, MDA fibriquick for fibrinogen determinations). We controlled the same lot reagent for each parameters during the period we studied. The reference ranges are 22-36 s for APTT, 11-14.2 s for PT, 180-520 mg/dL for fibrinogen. Total imprecision (CV) on the MDA II is comprised between 3.1-5.2 % for TriniCLOT PT HTF, 3.3-5.3 % for Platelin LS and 4.2-6.5 % for MDA fibriquick. Hexokinase method has been used as glucose reference technique. In this technique, precision calculations of the serum samples in mmoles per litres have been studied for all 3 levels and found as 2.3, 1.1 and 1.8 respectively. The numbers below the values have shown to symbolizing the sub-group number and % in the related case number more clearly.

***Statistical Analysis***

Statistical analysis was carried out using SPSS, (SPSS Base 15.0) version. Abnormal distribution of groups was detected by Kolmogorov Smirnov test. Results were finally reported as median and 5 th-95 th percentile distribution because results were not normally distributed. The significance of differences in parameters between groups was assessed by the Wilcoxon Mann-Whitney test. $p<0.05$ was considered as significant.

## Results

The age, sex, APTT, PT and fibrinogen values were compared after they were grouped according to FPG levels (Table 1). In our 1295 study cases with more than 7 mmol/L of glucose, 182 patients (14.1%) had APTT less than 22 s and 63 had (4.9%) had APTT values between 15.9-20.4 s. Comparison by Mann-Whitney test revealed shortened APTT in both IFG and diabetic groups compared to euglycemics ($p<0.001$, $p<0.001$ respectively) (Table 1). Statistically significant higher fibrinogen levels were also found for the same groups ($p<0.001$). APTT <22 s and fibrinogen *>520 mg/dL* were more frequently observed in the diabetic group (14.1% and 31.4%, respectively) than in the other two groups. No significant differences were found for age, gender and PT among classes. We have no data for clinical events in any of the groups because our study was a retrospective study.

## Discussion

A shortened APTT, either as a risk factor or as a marker for hipercoagulability has gained interest recently. Diabetic patients are characterized by significantly elevated levels of intrinsic pathway and fibrinogen, which are mainly determinants of the APTT (9). It has been shown that shortened APTTs may also reflect procoagulant imbalances with increased levels of coagulation factors. Therefore, APTT can be used to assess the risk of thromboembolic complications in diabetic patients (5, 12). Even though the shortened APTTs may be

**Table 1.** APTT, PT and fibrinogen levels of patients (n=5602), grouped according to FPG levels

| | FPG (mmol/L) | | |
|---|---|---|---|
| | < 5.6 | 5.6-6.9 | ≥7.0 |
| n | 2913 | 1394 | 1295 |
| Age | 47 (25-74) | 51 (27-72) | 55 (25-73) |
| Male % | 1387 (47%) | 612 (43%) | 602 (46%) |
| APTT (s) | 27 (22-31.3) | 26.2 (21.1-31.1)* | 25.1 (20.4-30.7)*, ** |
| <22 s (%) | 125 (4.3%) | 125 (9%) | 182 (14.1%) |
| PT (s) | 12.7 (11.4-14.1) | 12.7 (11.3-14.2) | 12.7 (11.4-14.2) |
| <11 s (%) | 29 (1%) | 19 (1.4%) | 28 (2.2%) |
| Fibrinogen mg/dL | 366 (223-677) | 396 (222-769) * | 440 (235-817)*, ** |
| >520 mg/dL | 448 (15.4%) | 376 (27%) | 406 (31.4%) |

* p< 0.001comparison with FPG< 5.6 mmol/L, ** p< 0.001comparison with FPG 5.6–6.9 mmol/L.

APTT; activated partial thromboplastin time, PT; prothrombin time, FPG; fasting plasma glucose levels.

as an independent risk factor for thromboembolic complications (5, 6), shortened APTTs may result from preanalytical causes such as inapporiate collection (13), handling and/or storage of the specimens (1, 14). Therefore preanalytical causes of shortened APTTs should be ruled out via repeating collection by an experienced phlebotomist and confirming the test. Then the possible clinically causes such as myocardial infarction (15), hyperthyroidism (16), cancer (17) should be considered for shortened APTTs.

It was reported that increased fibrinogen levels were a strong and independent cardiovascular risk factor (18). A positive correlation between plasma glucose and fibrinogen levels has been reported (19). Chronic hyperglycemia and glycation effect on fibrin structure and function, generating a clot with a denser structure which is more resistant to fibrinolysis (20). Because, fibrinogen may become hyperglycosylated in a hyperglycemic environment. The resulting fibrin structure is composed of small diameter fibers and they are markedly resistant to degradation (21). Moreover, hyperglycemia contributes to the hyperfibrinogenemia and activates the coagulative cascade in diabetics. Hepatic fibrinogen synthesis is substantially increased in normoalbuminuric type 2 diabetic patients (18).

There are several studies about shortened APTT and its potential value for different diseases such as myocardial infarction, venous thromboembolism (4, 5, 15). Reccently, Zhao et al. (11) reported that diabetic and high-risk diabetic patients have shortened APTTs and elevated fibrinogen levels. The APTT values were significantly shorter and fibrinogen values were significantly higher in the diabetic and IFG groups than in the normal groups. APTT values below the reference range and fibrinogen values above the reference range were more frequent in diabetics. They did not report any significant differences APTT and fibrinogen values between diabetic group and IFG group. Lippi et al (12) found shorter APTT values in the diabetic and IFG groups than in the euglycemic group. They also found APTT values below the reference range were significantly more frequent in diabetic group. No significant differences were reported by means of fibrinogen values. We think different results may result from FPG values which are the grouping parameters. Because the variability of FPG values with day-today within-person variance is 12–15% (22). Our study is retrospective analysis. We performed on patients, referred to our laboratory for routine blood testing. We did not determine variability of FPG within person. We could not find any differences for PT values between groups. Our results were concordance with literature findings (11, 12).

In our study, we did not measure the levels of single clotting factors. As thrombophilia tests are expensive and difficult to perform, shortened APTT can be used to identify procoagulant disequilibrium and it might allow the identification of a subset of patients at major risk of thromboembolic complications (12). Our study has some limitations. We did not measure platelet count and volume. Recently, it was reported that the levels of von Willebrand factor (vWf) and mean platelet volume (MPV) were significantly higher in the IFG group than the control group. The authors also suggested vWf levels were positively correlated with MPV levels in IFG group (23). In conclusion, we found significantly shortened APTT values and increased fibrinogen levels in both

diabetic group and IFG group when compared with the euglycemic group. Further studies are needed to confirm these findings.

## References


1. Langdell RD, Wagner RH, Brinkhous KM. Effect of antihemophilic factor on one-stage clotting tests: a presumptive test for hemophilia and a simple one-stage antihemophilic factor assay procedure. J Lab Clin Med 1953; 41:637–647.
2. Proctor RR, Rapaport SI. The partial thromboplastin time with kaolin: a simple screening test for first stage plasma clotting factor deficiencies. Am J Clin Path 1961; 36:212–219.
3. Ten Boekel E, Böck M, Vrielink GJ,et al. Detection of shortened activated partial thromboplastin times: an evaluation of different commercial reagents. Thromb Res 2007; 121:361–367.
4. Korte W, Clarke S, Lefkowitz JB. Short activated partial thromboplastin times are related to increased thrombin generation and an increased risk for thromboembolism. Am J Clin Pathol 2000; 113:123–127.
5. Tripodi A, Chantarangkul V, Martinelli I, et al. A shortened APTT is associated with the risk of venous thromboembolism.Blood2004; 104:3631-4.
6. Hron G, Eichinger S, Weltermann A, et al. Prediction of recurrent venous thromboembolism by the activated partial thromboplastin time. J Thromb Haemost 2006; 4:752–6.
7. Clement S, Braithwaite SS, Magee MF, et al. American Diabetes Association Diabetes in Hospitals Writing Committee Management of diabetes and hyperglycemia in hospitals. Diabetes Care 2004; 27:553-91.
8. Ceriello A. Coagulation activation in diabetes mellitus: the role of hyperglycaemia and therapeutic prospects. Diabetologia 1993; 36:1119-25.
9. Grant PJ. Diabetes mellitus as a prothrombotic condition. J Intern Med 2007; 262:157-72.
10. Reddy NM, Hall SW, MacKintosh FR. Partial thromboplastin time: prediction of adverse events and poor prognosis by low abnormal values. Arch Intern Med 1999; 159:2706–2710.
11. Zhao Y, Zhang J, Zhang J, Wu J. Diabetes mellitus is associated with shortened activated partial thromboplastin time and increased fibrinogen values. PLoS One 2011; 28;6(1):e16470.
12. Lippi G, Franchini M, Targher G, et al. Epidemiological association between fasting plasma glucose and shortened APTT. Clin Biochem 2009; 42:118–120.
13. Lippi G, Salvagno GL, Montagnana M, Guidi GC. Short-term venous stasis influences routine coagulation testing. Blood Coagul Fibrinolysis 2005; 16:453–458.
14. Lippi G, Favaloro EJ. Activated partial thromboplastin time: new tricks for an old dogma. Semin Thromb Hemost 2008; 34:604–611.
15. Madi AM, Greci LS, Nawaz H, et al. The activated partial thromboplastin time in early diagnosis of myocardial infarction. Blood Coagul Fibrinolysis 2001; 12:495–499.
16. Lippi G, Franchini M, Targher G, et al. Hyperthyroidism is associated with shortened APTT and increased fibrinogen values in a general population of unselected outpatients. J Thromb Thrombolysis 2009; 28:362–365.
17. Del Carpio-Cano FE, Zamora-Rangel D, Rosales-Manoatl AM, et al. Plasma circulating levels of soluble P-Selectin and Thrombospondin-1 (TSP1) are elevated in patients with cancer and a shortened activated partial thromboplastin time (APTT) without clinical evidence of thrombosis: an inexpensive and simple test (APTT) to be considered for its clinical value. Blood (ASH Annual Meeting Abstracts) 2006; 108;4098.
18. Barazzoni R, Zanetti M, Davanzo G, et al. Increased fibrinogen production in type 2 diabetic patients without detectable vascular complications: correlation with plasma glucagon concentrations. J Clin Endocrinol Metab 2000; 85:3121–3125.
19. Kannel WB, D'Agostino RB, Wilson PW, et al. Diabetes, fibrinogen and risk of cardiovascular disease: the Framingham experience. Am Heart J 1990; 120:672–676.
20. Franchini M, Targher G, Montagnana M, et al. The metabolic syndrome and the risk of arterial and venous thrombosis. Thromb Res 2008; 122:727-35.
21. Carr ME. Diabetes mellitus: a hypercoagulable state. J Diabetes Complications 2001; 15:44–54.
22. Mostafa SA, Davies MJ, Srinivasan BT, et al. Should glycated haemoglobin (HbA1c) be used to detect people with type 2 diabetes mellitus and impaired glucose regulation? Postgrad Med J 2010; 86:656–662.
23. Bostan F, Coban E. The relationship between levels of von Willebrand factor and mean platelet volume in subjects with isolated impaired fasting glucose. Med Sci Monit. 2011; 17(6):1-4.