ASSESSMENT OF THROMBOPHILIC STATUS IN TYPE 2 DIABETIC PATIENTS: THE LINK BETWEEN PAI-1 POLYMORPHISM AND THE ONSET OF COMPLICATIONS

Iris Bararu¹, M. Apavaloaie¹, O. Badulescu¹, C. Plesoianu², M. Ciocoiu¹, M. Badescu¹*

University of Medicine and Pharmacy „Grigore T. Popa”- Iași
Faculty of Medicine
1. Department of Morfo-functional Sciences
2. Department of Medical Specialties (I)
*Corresponding author. E-mail: magda.badescu@gmail.com

ASSESSMENT OF THROMBOPHILIC STATUS IN TYPE 2 DIABETIC PATIENTS: THE LINK BETWEEN PAI-1 POLYMORPHISM AND THE ONSET OF COMPLICATIONS (Abstract): Diabetes is a very common disease characterized by a prothrombotic status involving both the chronic activation of the clotting system and a decrease in the endogenous fibrinolytic capacity. The existing data regarding the hemostatic alterations in diabetic patients are insufficient and contradictory due to the heterogeneity of the studied population, the presence of cardiovascular complications, glycemic status, other associated risk factors, and the type of treatment. The metabolic changes found in diabetic patients cause disturbances in platelet activity, hemostasis, endogenous fibrinolysis and rheology favoring a prothrombotic status. These systemic changes may lead to an extensive atherosclerotic process derived from the thrombophilic status, thus partially explaining the frequent coronary atherosclerosis in type 2 diabetic patients. Moreover, the hemostatic alterations seen in diabetic patients seem to be responsible for the microvascular localization of coronary atherosclerosis. Recent studies indicate that increased PAI-1 levels are correlated with an increase in the incidence of diabetes mellitus and its complications, such as diabetic neuropathy, diabetic retinopathy and ischemic cardiomyopathy. PAI-1 is synthesized both by endothelial cells and hepatocytes. Endothelial cell activation causes an increase in tissue thromboplastin secretion, a decrease in thrombomodulin expression, a diminished tissue plasminogen activator synthesis and an increase in PAI-1 production. Despite all this, the data regarding the association between PAI-1 levels and the incidence of diabetes mellitus remain unclear. Keywords: TYPE 2 DIABETES MELLITUS, THROMBOPHILIC STATUS, PAI-1 POLYMORPHISM.

Diabetes is a very common disease that is characterized by a prothrombotic status involving both the chronic activation of the coagulation system and a decrease in endogenous fibrinolytic capacity. The studies comparing the prevalence of coronary diseases in diabetic and nondiabetic patients showed a threefold higher incidence of atherosclerosis, with an earlier clinical onset and a twofold higher cardiovascular risk in diabetic patients.

Hyperlipoproteinemia, platelet activation and changes in fibrinogen levels seem to play an important role in the onset and progression of diabetic complications. The existing data regarding the hemostatic
changes in diabetic patients are insufficient and inconclusive due to the heterogeneity of the studied population, presence or absence of cardiovascular complications, glycemic status, other associated risk factors, and the type of administered treatment received (1).

It is well known that the vascular diseases in diabetic patients have a multifactorial etiology, being influenced by the changes in thrombosis and thrombolysis balance, consisting mostly in the activation of the thrombotic process, thus leading to ischemic complications.

The association of increased LDL-cholesterol levels, abnormal triglyceride levels, small and dense LDL-cholesterol particles and diminished HDL-cholesterol levels leads to the atherogen lipoprotein phenotype, which is correlated with an early onset of coronary disease and extensive atherosclerosis process in type 2 diabetic patients. Moreover, small and dense LDL particles seem to be a marker for a series of anomalies including the decrease of HDL-cholesterol levels, the increase in apoB concentrations, diminished insulin sensitivity and procoagulant anomalies (increased PAI-1 levels) (2). The association between the atherogenic lipoprotein phenotype and other proatherogenic anomalies is suggestive of a genetic predisposition. Therefore, in 1990 it was proven that the genetic locus responsible for small and dense LDL particles is also associated with increased triglyceride levels, apoB, VLDL and IDL levels and decreased HDL-cholesterol and apoA1 levels. Even more, genetic linkage studies showed that LDL particle size and HDL and LDL cholesteral levels are correlated with an allele of hepatic lipase gene. The genetic mutations affecting the Cholesteryl Ester Transfer Protein (CETP) locus situated on the 16th chromosome can influence, through its role in the reverse transport of cholesterol, the LDL size and triglyceride levels (3).Furthermore, there is a constant relation between the atherogenic lipoproteins (oxidized LDL and LP(a)) and the vascular endothelium, thus leading to a diminished fibrinolytic process. The effect is an imbalance between coagulation and fibrinolysis that favors thrombosis which eventually causes the occurrence of thrombotic complications (4,5). Hypercoagulability in type 2 diabetes mellitus seems to result especially from the synthesis of factors that activate the coagulation system and factors that inhibit fibrinolysis (such as factor VII or PAI-1). The hemostatic anomalies in type 2 diabetic patients may also reside from the synthesis in the adipose tissue of inflammatory cytokines that can also mediate insulin resistance, such as IL-6 and TNF-alpha (6).

**ASSESSING THE PROTHROMBOTIC STATUS IN TYPE 2 DIABETES MELLITUS PATIENTS**

Recent studies showed that both endogenous fibrinolytic parameters and platelet activity are changed in type 2 diabetic patients at increased cardiovascular risk. The metabolic alterations that cause a change in rheology, platelet activity, hemostasis and endogenous fibrinolysis found in diabetic patients favor the occurrence of a thrombophilic status. These changes can cause an extensive atherosclerotic process derived from the thrombophilic status, thus partially explaining the frequent coronary atherosclerosis in type 2 diabetic patients. Even more, the hemostatic alterations seen in diabetic patients seem to be responsible for the microvascular localization of coronary atherosclerosis (7, 8).
Assessment of thrombophilic status in type 2 diabetic patients: the link between Pai-1 polymorphism and the onset of complications

The plasminogen activator inhibitor-1 (PAI-1) belongs to the serine protease inhibitor superfamily, and plays a key role in the regulation of extracellular matrix degradation. Studies have indicated that the increase in PAI-1 level was related to the incidence of diabetes mellitus (DM) and its complications, such as an increased risk of diabetic nephropathy (DN), diabetic retinopathy (DR) and diabetic coronary artery disease (CAD). PAI-1 is synthesized by the endothelial cells and hepatocytes. Endothelial cell activation determines an increase in tissue thromboplastin secretion, a decrease in thrombomodulin expression and an increase in PAI-1 synthesis. Some studies indicated the presence of elevated PAI-1 serum levels in diabetic patients (9). Structurally modified lipoproteins such as VLDL or LDL have the capacity of releasing PAI-1, thus causing a decrease in the fibrinolytic plasmatic capacity, determined by a diminished conversion of plasminogen to plasmin. There are 5 identified polymorphisms of PAI-1 gene: -675 4G/5G, -844 A>G, Ala15Thr, Val17Ile and Asn195Ile; the presence of these polymorphisms may increase the cardiovascular risk in diabetic patients (10). The PAI-1 gene is situated on the 7th chromosome and contains 8 introns and 9 exons. The 4G/5G polymorphism (the deletion/insertion of guanosine in the 675 position of the PAI-1 gene promoter) is a major genetic determinant of PAI-1 levels. Some authors suggested that the association of the 4G allele with other thrombophilic defects (such as factor V Leiden, prothrombin gene mutation, hyperhomocysteinemia, diminished protein C and protein S activity) increases the risk of thrombosis. In diabetic patients with coronary ischemic disease the presence of 4G/4G genotype was associated with an increased risk of sudden death.

LESSONS FROM CLINICAL TRIALS

Some of the oldest studies that evaluated the association between PAI-1 polymorphism and the risk of type 2 diabetes mellitus showed a positive correlation between the 4G allele of type 1 plasminogen activator and the risk of diabetes mellitus in Pima Indians, in this population the incidence of noninsulin-dependent diabetes being very high. On the contrary, some subsequent studies analyzing the Caucasian population did not manage to show an association between PAI-1 polymorphism and diabetes mellitus. A case-control study on 856 Tunisian patients with type 2 diabetes mellitus showed an increased incidence of 4G/4G genotype, thus suggesting that this polymorphism may be a prothrombotic risk factor associated with type 2 diabetes mellitus. Many studies analyzed the incidence of PAI-1 polymorphism in diabetic Asian patients, but did not succeed to prove a significant statistic correlation between 4G/5G allele and the risk of diabetes (6).

A meta-analysis published in 2013 analyzed the results of 9 studies including 1219 patients, from which 5 studies referred to Caucasian population, 3 studies included Asian patients and 1 study was conducted on Pima Indians. This meta-analysis demonstrated a slight significant statistic correlation between 4G/4G polymorphism and the risk of diabetes complicated with neuropathy. In the subgroup analysis by ethnicity, significantly increased risks were observed among the Caucasian population (for 4G/4G versus 5G/5G, for 4G/4G versus 5G/5G + 4G/5G). In the stratified analysis by average diabetes duration, PAI-1 variation was found associated with a high risk of diabetes retinopathy in patients with duration of diabetes...
tes longer than 10 years (for 4G/4G versus 5G/5G).

Another meta-analysis conducted in 2013, consisting of 20 studies, tried to assess the relationship between PAI-1 polymorphism and the risk of developing diabetes mellitus and its complications. Therefore, 6 studies including 1333 patients analyzed the association between PAI-1 675 4G/5G polymorphism and the risk of diabetes, 7 studies including 1060 patients assessed the correlation between this polymorphism and diabetic nephropathy, 10 studies with 1327 patients evaluated the correlation with diabetic retinopathy and 4 studies with 610 patients measured the risk of coronary artery disease in type 2 diabetic patients. The results of this meta-analysis suggested that there is no conclusive association between PAI-1 4G allele and the risk of diabetes mellitus or of developing complications, no matter the type of the evaluated population or diabetes duration.

CONCLUSIONS

The existing data are rather contradictory, some studies suggesting that high PAI-1 levels are constantly present in patients with type 2 diabetes mellitus or with insulin resistance, other studies denying this hypothesis. PAI-1 675 4G/5G polymorphism determines the deletion/insertion of a single guanosine, thus causing the expression of an additional linkage DNA situs for a protein with a repressing role in the transcription process, which will have an increased role in PAI-1 plasma levels. Therefore, this polymorphism may become a genetic risk factor for diabetes mellitus. The elevated PAI-1 levels may be correlated with a high incidence of such diabetic complications as diabetic nephropathy, diabetic retinopathy and coronary artery disease, but the results of the existing trials remain unclear and inconclusive. In these conditions unrevealing new genes that contribute to the thrombophillic status present in diabetic patients remains an important fundamental research theme.

ACKNOWLEDGMENTS

This paper was published under the frame of European Social Found, Human Resources Development Operational Programme 2007-2013, project no. POSDRU/159/1.5/S/136893.

REFERENCES

Assessment of thrombophilic status in type 2 diabetic patients: the link between Pai-1 polymorphism and the onset of complications


