High risk haemostasis patterns in overweight patients with type 2 diabetes mellitus

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PATRÓN DE HEMOSTASIA DE ALTO RIESGO EN PACIENTES CON SOBREPESO Y DIABETES TIPO 2

Introducción. Algunos estudios han demostrado un estado de hipercoagulabilidad en los pacientes diabéticos.

Objetivo. Comparar el patrón de hemostasia entre pacientes diabéticos con sobrepeso y controles.

Diseño. Un total de 23 pacientes con diabetes mellitus tipo 2 y sobrepeso atendidos en nuestra unidad fueron incluidos en el estudio (16 varones/7 mujeres). Las características clínicas de los pacientes fueron: 61,3 ± 12,3 años, índice de masa corporal (IMC) 27,2 ± 3,9 kg/m² y duración de la diabetes 8,4 ± 6,7 años. Un grupo de 23 voluntarios sanos fue elegido al azar entre donantes de sangre sin diabetes mellitus (15 varones/8 mujeres). Las características clínicas de estos pacientes fueron: 62 ± 13 años e IMC 27,6 ± 3,1 kg/m². A todos los sujetos, casos y controles, se les realizaron las siguientes pruebas: Hemoglobina glucosilada (HbA1c), parámetros de hipercoagulabilidad (t-PA, antígeno Von Willebrand (vW), proteína C (PC), proteína S (PS), trombomodulina (TH), factor VII activado, dimero D (DD), plasmina-antiplasmina (PAP) y fragmento activado protrombina F1 + 2 (F12). Estos parámetros fueron comparados en ambos grupos, y dentro de los diabéticos en los grupos con y sin micro y macroangiopatía. En ambos grupos, se realizó un análisis de correlación entre los parámetros clínicos y los hemostásicos.

Resultados. Los pacientes diabéticos con sobrepeso evidenciaron un incremento en los factores procoagulantes (F12 1,38 ± 0,4 frente a 1,21 ± 0,25 mmol/l; p < 0,05; VIgM 94,6 ± 48 frente a 91,7 ± 28 (MU/ml); p < 0,05) y, a la vez, una disminución en los parámetros fibrinolíticos (TAT, 74,5 ± 143 frente a 107,5 ± 248,5 µg/l; p < 0,05) y anticoagulantes (TAT, 117 vs 4,1 ng/ml; p < 0,05). En diabéticos, los parámetros con presencia de micro y macroangiopatía no hubo diferencia en función de la ausencia o presencia de micro o macroangiopatía. La proteína C y tPA mostraron una correlación negativa (-0,32; p < 0,05, respectivamente) con la hemoglobina glucosilada (HbA1c). FvW se correlacionó de una manera positiva con el IMC (r = 0,32; p < 0,05). No se encontraron correlaciones entre los parámetros de hemostasia, con el IMC y la HbA1c, en los sujetos control.

Conclusion. En los pacientes con diabetes tipo 2, hay un estado de hipercoagulabilidad que puede influir en las complicaciones crónicas de esta población.

Palabras clave: Diabetes mellitus tipo 2, Hipercoagulabilidad, Sobrepeso.

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INTRODUCTION

Diabetes mellitus is an independent risk factor for the development of atherosclerosis. The possible mechanisms are unclear. It is postulated that chronic inflammation may contribute to the increase of the risk of coronary heart disease in different ways: increasing serum concentrations of acute phase reactants (such as fibrinogen or C reactive protein) or modifying the serum lipid pattern (such as decrease of HDL-cholesterol and increase of triglycerides). Another factor involved in the atherogenesis of diabetic patients is the promotion of the oxidation of LDL-cholesterol since oxidation enhances the atherogenic capacity of those molecules. More recently, some authors have shown that diabetic patients had a hypercoagulable state.

The aim of our study was to compare the haemostasis pattern between overweight patients with diabetes mellitus type 2 and a control group.

MATERIALS AND METHODS

Population

Twenty-three overweight patients of our Diabetes Unit (16 males/7 females) and twenty-three controls (blood donors) (15 males/8 females) without diabetes were studied. Both groups were chosen at random and their characteristics are shown in table 1. Patients and controls did not take either anti-hypertensive or hypolipemic drugs. Diabetic patients took sulfonyleureas as antihyperglycemic agents. The study was approved by the local ethical committee and each patient gave informed consent to participate in the study.

Design

All patients (diabetic and controls) underwent the following examinations: plasma triglyceride activator inhibitor type (TAP)-1, thrombin-antithrombin III complex (TAT), tissue plasminogen activator (t-PA), von Willebrand factor (vWF), protein C (PC), protein S (PS), thrombomodulin (TH), activated VII factor, D dimer (DD), plasminogen activator inhibitor type 1(PAI-1), thrombin/antithrombin III complex (TAT), tissue plasminogen activator fragment F1+2 (F12). Blood samples for coagulation testing were collected into 3.1 g/L trisodium citrate solution between 07:00 and 09:00 am, after the subjects had fasted for 12 h. Samples were centrifugated for 15 min at 250 × g at room temperature.

Haemostasis assessment

Plasminogen activator inhibitor type I (PAI-1) (normal range < 10 U/ml) and tissue plasminogen activator (t-PA) (normal range 1-12 ng/ml) were determined by enzyme immunoassay (TintElize ELISA kit, USA). Thrombin/antithrombin III complex (TAT) (normal range 1.94-37.7 ng/L) was determined by enzyme immunoassay (Tinaquant TAT II HIAC; Marburg, Germany). Von Willebrand factor (vWF) (normal range 50-160%) was determined by enzyme immunoassay (American Diagnostica, USA). Protein C and S were determined by coagulometry, (normal range 70-150% for both). Thrombomodulin (normal range 14-55 ng/ml), and plasmin-antiplasmin (PAP) (normal range 99-368 ng/L) were determined by enzyme immunoassay (Tinaquant TAT II HIAC, Marburg, Germany). Factor VII (a) (normal range 5-85 μU/ml) was determined by coagulometry (Staclot, Asnieres-Seine, France). Factor VIII activity with a commercial kit (Enzygnost TAT micro, Marburg, Germany). Thrombin (normal range 17-76 ng/L), prothrombin activation fragment F1+2 (F12), plasminogen activator inhibitor type 1(PAI-1) (normal range 0-10 U/ml) von Willebrand antigen (vW), protein C (PC), protein S and protein S (PS), thrombomodulin (TH), activated VII factor, D dimer (DD), plasminogen activator inhibitor type 1(PAI-1), thrombin/antithrombin III complex (TAT), tissue plasminogen activator fragment F1+2 (F12), blood samples for coagulation testing were collected into 3.1 g/L trisodium citrate solution between 07:00 and 09:00 am, after the subjects had fasted for 12 h. Samples were centrifugated for 15 min at 250 × g at room temperature.

TABLE 1. Clinical characteristics of overweight diabetic patients and controls

| Age (years) | 63.1 ± 12.3 | 62.1 ± 13 | NS |
| Sex (male/female) | 16/7 | 15/8 | NS |
| BMI (kg/m²) | 27.2 ± 3.9 | 27.6 ± 3.1 | NS |
| Diabetes duration (years) | 8.4 ± 6.7 | 6.3 ± 3.1 | NS |
| HbA1c (%) | 8.9 ± 2.6 | 8.9 ± 2.4 | NS |
| Microangiopathy (%) | 12.8% | 0 | NS |
| Macroangiopathy (%) | 46.9% | 0 | NS |

Peripheral vascular disease was clinically defined by the presence of intermittent claudication, absent or weakened peripheral pulses, or both. Retinopathy was documented by standard fundus eye examination and diagnosed on the presence of microaneurysms, venous dilation, cotton-wool spots, neovascularization or hemorrhages. Clinical neuropathy was defined by an abnormal neurologic examination, consistent with the presence of peripheral sensorimotor neuropathy. Nephropathy was defined by the presence of urinary albumin excretion of 30 mg or more per 24 hours.

TABLE 2. Differences in hemostasis parameters in diabetic and control patients

<table>
<thead>
<tr>
<th>Procoagulant parameters</th>
<th>Diabetic patients (n = 23)</th>
<th>Control subjects (n = 23)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>AP-1 (U/ml)</td>
<td>1.38 ± 0.4</td>
<td>1.21 ± 0.25</td>
<td>&lt; 0.05</td>
</tr>
<tr>
<td>PAI (U/ml)</td>
<td>25.3 ± 24.9</td>
<td>37.3 ± 51.7</td>
<td>NS</td>
</tr>
<tr>
<td>TAT (μg/L)</td>
<td>5.52 ± 2.8</td>
<td>5.63 ± 4.5</td>
<td>NS</td>
</tr>
<tr>
<td>VII (μM/mL)</td>
<td>94.6 ± 48</td>
<td>81.7 ± 28</td>
<td>&lt; 0.05</td>
</tr>
</tbody>
</table>

Fibrinolytic parameters

| PAP(ng/l) | 262.9 ± 107.5 | 348.5 ± 143 | < 0.05 |
| t-PA (ng/ml) | 12.6 ± 5.1 | 7.4 ± 3.1 | < 0.05 |
| FvW (%) | 188 ± 57.1 | 119 ± 27.4 | NS |
| DD (μg/l) | 22.3 ± 26.8 | 9 ± 5.4 | < 0.05 |

Anticoagulant parameters

| Protein C (%) | 106 ± 32 | 120.7 ± 23 | NS |
| Protein S (%) | 97.7 ± 24 | 98 ± 25 | NS |
| Thrombomodulin | 27.4 ± 11.7 | 45.1 ± 21.7 | < 0.05 |
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a significant increase in prothrombin activation fragment F1+2 and factor VII(a). Values of PAI and TAT did not have statistical differences. Fibronectin parameters showed significant differences in TPA and D dimer (increased) and PAP (decreased), without differences in FvW. A decrease in anticoagulant parameters was observed in diabetic patients (thrombomodulin), without differences in protein S and C.

All haemostasis parameters were compared in diabetic patients in the group with (BMI > 25.9 kg/m²) and without microangiopathy (51.1%), but no differences were found. In a correlation analysis between HbA1c and haemostatic parameters, only protein C and TPA showed significant inverse correlations (r = –0.34; p < 0.01 and r = –0.32; p < 0.05, respectively). Another correlation analysis was performed between BMI and haemostasis parameters, only FvW was correlated with BMI (r = 0.32; p < 0.05). No correlations were found between haemostasis parameters with BMI and HbA1c in non-diabetic subjects.

No correlations were found among diabetes evolution, age or microalbuminuria levels with haemostasis parameters in diabetic patients.

DISCUSSION

Patients with type 2 diabetes mellitus have a variety of coagulation dysfunctions, which could contribute to microvascular and macrovascular complications. The hypercoagulable state has been demonstrated in a group of overweight diabetic patients under strict metabolic control who had an increase in TAT levels. In our study no significant differences in TAT levels were detected between overweight diabetic and control subjects, but F1+2 and activated factor VII were increased in diabetic patients, showing a hypercoagulable state. In diabetic patients, it has been shown that antithrombotic parameters, such as PAI-1 and t-PA antigen, were strongly related to insulin resistance, whereas the link with factor VII and other procoagulant parameters remained weak. These alterations might contribute to increase cardiovascular mortality in diabetes. For example, Morishita et al. showed significantly higher levels of TAT, fibrinogen and PAI-1 in 22 diabetics patients with coronary heart disease than 51 patients without diabetic macroangiopathy.

Another haemostasis alteration in overweight diabetic patients is a decrease in the anticoagulant system. Patients with diabetes have activated protein C resistance, suggesting that final steps of the protein C/S inhibiting system might be altered in relatives of type 2 diabetic patients, who in a case-control study exhibited levels of prothrombin F1+2 and D dimer than control subjects.

An additional point of interest is the relationship between some haemostasis parameters and BMI. One possibility is that changes in these parameters are related to adipose tissue derived cytokines.

In conclusion, hypercoagulable state is present in diabetic patients which with present knowledge can be viewed as a risk factor for chronic complications. The role of adipose tissue as a possible cause of chronic inflammatory activity in diabetic patients requires further investigation.

REFERENCES


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