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Letter to the Editor

Granulocyte cytosolic calcium in type 2 diabetes

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Diabetes mellitus (DM) is associated with a granulocyte dysfunction, more evident evaluating specific functions such as adherence, chemotaxis, phagocytosis, bactericidal and oxidative activity. Many of these functions reveal their impairment especially after *in vitro* activation.

The granulocyte dysfunction is related to the metabolic alteration that characterizes DM. This alteration regards the reduced activity of phosphofructokinase and so the glycolytic pathway, the increase of glucose metabolism via the hexose monophosphate shunt and the activation of the polyol pathway and so the increased concentration of cell sorbitol.

In addition to the dysfunction, a spontaneous activation of diabetic granulocytes is shown by an increase in the plasma concentration of elastase [1,2].

A possible marker of granulocyte dysfunction/activation is the cytosolic Ca^{2+} concentration change [3]. Indeed, in DM the intracellular calcium content increases in several tissues and this increase is often tissue specific [4].

The granulocyte cytosolic calcium is dependent on membrane pumps [5] and their activity besides to be influenced by the level of blood glucose or insulin or both, is also dependent on the membrane fluidity [6], that is a component of the granulocyte deformability usually reduced in DM [7]. This reduction may lead to the impairment of circulation and plays a role in the pathogenesis of diabetic microangiopathy.

Previously, examining the granulocyte cytosolic Ca^{2+} concentration in different groups of diabetic subjects of type 1 and 2, we observed that this parameter was not discriminant in comparison with normal controls [8,9]. Here we have evaluated the same parameter in a larger group of type 2 diabetics subdivided according to the presence or absence of macrovascular complications, being the latter evaluated both instrumentally (Doppler, echo-Doppler, ECG, etc.) and by physical examination.

We included 54 type 2 diabetics (24 men and 30 women; mean age 58.0 ± 8.6 years) without macrovascular complications (MVC) and 50 type 2 diabetics (36 men and 14 women; mean age 61.7 ± 8.5 years) with MVC. In the group of type 2 diabetics without MVC the fasting blood glucose level was 10.32 ± 3.44 mmol/l, the total cholesterol level was 5.55 ± 0.92 mmol/l and serum triglycerides were 1.49 ± 0.60 mmol/l. In the group of type 2 diabetics with MVC the fasting blood glucose level was 9.17 ± 3.14 mmol/l, the total cholesterol level was 5.91 ± 1.22 mmol/l and serum triglycerides were

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2.27 ± 1.01 mmol/l. All the type 2 diabetics followed a controlled carbohydrate diet and received oral hypoglycaemic agents.

The granulocyte Ca^{2+} concentration was evaluated marking the cells with the fluorescent probe Fura 2-AM and the ratio between the Fura 2–Ca^{2+} complex and the fluorescence intensity of the unchelated Fura 2 was evaluated [10].

The granulocyte Ca^{2+} concentration was also examined in a group of 39 normal controls (25 men and 14 women; mean age 38.8 ± 7.0 years). In this group the fasting blood glucose level was 5.08 ± 0.49 mmol/l, the total cholesterol level was 5.09 ± 0.92 mmol/l and the serum triglycerides were 1.16 ± 0.64 mmol/l.

In normal controls and in type 2 diabetics without and with MVC no statistical correlation was found between age and granulocyte cytosolic Ca^{2+} content.

No significant difference was observed in granulocyte cytosolic Ca^{2+} concentration between normal controls and type 2 diabetics without MVC (N $= 0.806 \pm 0.056$; DM without MVC $= 0.821 \pm 0.070$) while an increase of this parameter was present in type 2 diabetics with MVC compared to normal controls (DM with MVC $= 0.865 \pm 0.031$; $p < 0.001$ vs N). The cytosolic Ca^{2+} concentration was however able to distinguish type 2 diabetics with and without MVC ($p < 0.001$).

These findings, different from that reported by other authors [11,12] who found an increase of this granulocyte variable in type 2 diabetics not subdivided for macrovascular complications, agree with our previous results [8,9] and in particular underline how in our experience only the presence of MVC is associated with a significant increase of granulocyte cytosolic Ca^{2+} content. In type 2 DM, especially complicated by macrovascular involvement, a pharmacological modulation of granulocyte cytosolic Ca^{2+} concentration by using calcium channel blockers, may be considered.

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