Glycemic Control Influences Serum Angiogenin Concentrations in Patients With Type 2 Diabetes

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OBJECTIVE — Because diabetes is the most frequent factor responsible for microvascular and macrovascular disease, we investigated angiogenin serum levels within the diabetic patient group.

RESEARCH DESIGN AND METHODS — We investigated 49 patients who met the criteria to be in the diabetic group. Forty nondiabetic patients were included in the control group. We set A1C <7% as well-controlled diabetes. Serum angiogenin level was measured using the enzyme-linked immunosorbent assay method.

RESULTS — Serum angiogenin levels of poorly controlled patients with type 2 diabetes were significantly lower than those of group with well-controlled diabetes (361.23 ± 126.03 ng/ml vs. 446.37 ± 134.10 ng/ml; $P = 0.001$). Moreover, they were characterized by a significantly longer duration of the disease ($P = 0.006$), higher BMI ($P = 0.0003$), and higher systolic blood pressure ($P = 0.01$). Levels of total cholesterol, triglycerides, LDL, and HDL were not significantly different in both groups.

CONCLUSIONS — Patients with poorly controlled type 2 diabetes (A1C >7%) have lower angiogenin levels than patients with well-controlled diabetes.

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Diabetes is associated with angiopathy, which increases the risk of a variety of complications. To minimize the degree of ischemic damage, collateral vessels of the large arteries are developed. This process is, unfortunately, insufficient in diabetic patients, leading to a three- to fourfold increase in mortality risk. Both coronary vessel formation and capillary density are poorer in patients with diabetes than in a healthy control group (1,2).

Angiogenesis is a complex process regulated by stimulatory and inhibitory factors. It is well established in research that the expression of different angiogenic growth factors are reduced in diabetes. On the other hand, diabetic patients with coronary artery disease exhibit higher levels of angiostatin and endostatin (3). In the meantime, the molecular angiogenic signaling pathways influencing coronary collateral formation remain poorly understood.

We have previously demonstrated that the serum level of one of the angiogenic growth factors, namely angiogenin, is reduced in diabetes (4). The present investigation was designed to test the hypothesis that the serum level of angiogenin depends on glucose concentration control.

RESEARCH DESIGN AND METHODS

— A total of 49 consecutive patients with type 2 diabetes from the Department of Family Medicine, Medical University of Gdansk, admitted to the Department of Ophtalmology for a routine control were recruited between September and December 2009. They were divided into two groups according to the level of A1C (18 subjects with A1C <7% and 31 subjects with A1C ≥7%). Patients suffering from acute and chronic infections and neoplastic diseases were excluded. Diabetes was defined according to American Diabetes Association criteria (5). All patients had blood glucose >7 mmol/l, and we set A1C <7% as well-controlled diabetes. Serum angiogenin levels were measured twice with the ELISA Quantkine kit (R&D Systems, Minneapolis, MN). A1C was evaluated by the immunoturbidimetric method using the Unimate 3 set (Hoffmann-La Roche, Basel, Switzerland). A group of 40 healthy age- and sex-matched individuals was used as a control. They remained under continuous supervision at The Family Medicine Outpatient Clinic.

The results were analyzed using Statistica software, version 8 (Stat Soft Polska). The level of significance was set at $P < 0.05$. Student t and the Kruskal-Wallis ANOVA tests were performed as standard.

RESULTS

— Serum angiogenin levels of poorly controlled patients with type 2 diabetes were significantly lower than those of the group with well-controlled diabetes ($P = 0.001$). Moreover, they were characterized by a significantly longer duration of the disease and higher BMI and systolic blood pressure. Levels of total cholesterol, triglycerides, LDL, and HDL were not significantly different from those of the well-controlled patients (Table 1).

The patients with A1C >7% suffered significantly more often from hypertension ($P = 0.003$) and ischemic heart disease ($P = 0.004$). However, there was no significant difference in the occurrence of late complications of diabetes.

There were significant differences between treatment regimes. The well-controlled patients received sulfonylurea significantly more often ($P = 0.0007$), while the poorly controlled group was treated mainly with insulin ($P = 0.002$). No significant differences in statin and ACE inhibitor treatment were observed.

The serum angiogenin level of the healthy control group was significantly higher than that of the total diabetic pa-

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Table 1—*Basic parameters of well-controlled and poorly controlled diabetic groups*

Parameter	A1C < 7%	A1C ≥ 7%	*P* value
n	18	31	
Age (years)	66.22 ± 12.34	66.29 ± 8.91	0.8436
Weight (kg)	75.18 ± 12.73	86.82 ± 16.15	0.0127
BMI (kg/m^2)	26.56 ± 3.37	32.05 ± 6.07	0.0003
Systolic blood pressure (mmHg)	130.53 ± 12.76	142.77 ± 16.87	0.0127
Diastolic blood pressure (mmHg)	72.41 ± 18.00	79.30 ± 9.26	0.1054
Diabetes duration (years)	7.12 ± 5.53	12.33 ± 6.33	0.0068
A1C (%)	6.17 ± 0.44	8.69 ± 1.40	0.0000001
Angiogenin (ng/ml)	446.37 ± 134.10	361.23 ± 126.03	0.0308
Total cholesterol (mmol/l)	195.18 ± 29.25	212.07 ± 38.09	0.1263
LDL cholesterol (mmol/l)	118.81 ± 23.87	127.48 ± 35.80	0.3948
HDL cholesterol (mmol/l)	45.82 ± 11.04	51.59 ± 20.23	0.3,467
Triglycerides (mmol/l)	145.56 ± 68.53	167.11 ±64.20	0.3055
Hypertension (%)	66.67	96.77	0.0037
Nephropathy (%)	5.56	6.45	0.8996
Retinopathy (%)	17.65	30.00	0.3507
Foot ulceration (%)	11.11	16.13	0.6285
Ischemic heart disease (%)	11.11	51.61	0.0046
ACE inhibitor (%)	52.94	80.00	0.0513
Statins (%)	29.41	51.72	0.1406
Sulphonylurea (%)	84.62	11.11	0.0007
Metformin (%)	53.85	22.22	0.1380
Insulin (%)	23.08	88.89	0.0024

Data are arithmetic means ± SD or percentage of patients.

tients (472.57 ± 146.30 ng/ml vs. 392.5 ± 134.2 ng/ml; $P = 0.000017$). ANOVA and post hoc NIR Fisher test detected significant differences between healthy subjects and patients with A1C >7% (472.57 ± 146.30 ng/ml vs. 361.23 ± 126.03 ng/ml; $P = 0.00002$) and within the two diabetic groups $P = 0.03$.

CONCLUSIONS — The rising prevalence of diabetes is leading to a significant increase in long-term diabetes complications such as retinopathy, nephropathy, neuropathy, and ischemic heart disease, which have a considerable impact on the patients (6). The data available on the effect of hyperglycemia on the process of angiogenesis are still limited. Numerous authors have suggested that a high concentration of glucose causes endothelial cell dysfunction (7,8). Weihrauch et al. (9) indicated that the production of inhibitors of angiogenesis is an important mechanism for the impairment of collateral development observed during hyperglycemia.

Angiogenin is an important and still poorly investigated angiogenic growth factor (10) that has a great influence on the process of creation of new vessels. When introduced into Balb/c mice in a serum-induced angiogenesis test, the serum angiogenin of diabetic patients causes a significant decrease in new vessel formation (4).

The present results confirm the findings of our previous studies and indicate the correlation between the glycemic control and the inhibition of the serum angiogenin level. We discovered that the serum angiogenin level is markedly decreased in patients with A1C >7% in comparison with well-controlled patients and their healthy counterparts. Furthermore, poorly controlled patients had a higher proportion of hypertension and ischemic heart disease and greater BMI. These data are comparable with the results of different observational studies (11). The longer the duration of diabetes, the poorer its control and the lower the serum angiogenin level.

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