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Diabetes mellitus and Alzheimer's disease

Dear Sir,

Recently in a letter in *Diabetologia* [1] it was suggested that “the occurrence of AGE (advanced glycation end products) in both diabetes and Alzheimer's disease might lead one to predict a higher-than-expected incidence of concurrent diabetes and Alzheimer's disease. However, predictions of diabetes and concurrent Alzheimer's disease unfortunately must await epidemiological analyses . . .” “Conversely, life-long glycation may result in compensations, leading instead to discordance of Alzheimer's disease and diabetes”.

The Umeå Dementia Research Group reported a population-based study of 839 patients with various dementia diagnoses of whom 457 were classified as multi-infarct dementia (MID), 317 as dementia of Alzheimer type (SDAT) and 65 as confusional states [2]. Sixty patients were found to have diabetes, none in the SDAT group, but 55 in the MID group and 5 in the group of confusional states. Oral glucose tolerance tests were performed in patients not known to have diabetes and

compared in patients with SDAT, MID, cerebrovascular diseases, hospitalized control patients and healthy elderly patients. Fasting blood glucose levels were lower and the areas under the oral glucose tolerance curves smaller in the SDAT group compared with the cerebrovascular and the hospitalized control groups. SDAT patients had higher insulin levels than the healthy elderly group. Our findings may be in accordance with the second suggestion mentioned above. However, initially low concentrations of brain transmitters are further negatively influenced by the lack of an important source of energy or perhaps by the lack of a metabolite necessary for the production of transmitters.

Yours sincerely,
F. Lithner

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Elevated circulating adhesion molecules in NIDDM – potential mediators in diabetic macroangiopathy

Dear Sir,

Increased expression of endothelial adhesion molecules (AMs) may have a potential role in accelerated diabetic macroangiopathy [1], possibly stimulated by oxidized low-density lipoproteins (LDL) and free radicals derived from glucose auto-oxidation [2], since “classical” risk factors including hyperlipidaemia, hypertension and obesity do not completely account for the increased incidence of atherosclerosis in diabetes [3]. Such adhesion molecules are involved in chemotaxis of cir-

culating monocytes and binding of leukocytes and platelets to the endothelium. This leukocyte recruitment is considered an initial step in development of atherosclerotic lesions [4]. Raised plasma concentrations of soluble circulating adhesion molecules occurring after proteolytic cleavage of the extracellular domain may indicate endothelial activation and increased interaction of leukocytes and platelets with the endothelium as recently shown for a small number of non-insulin-dependent diabetic (NIDDM) patients ($n = 13$) [5] and diabetic patients not requiring insulin treatment [6].

In a cross-sectional study, we therefore analysed serum concentrations of circulating intercellular adhesion molecule-1 (cICAM-1), vascular cell adhesion molecule-1 (cVCAM-1), and endothelial leukocyte adhesion molecule-1 (cELAM-1 = E-selectin) in a large group of metabolically stable NIDDM patients ($n = 159$) and age-matched healthy subjects ($n = 71$) with no clinically manifest infections and who were not receiving any anti-inflammatory medication. NIDDM patients were treated either by diet alone, with additional sulphonylurea or biguanides ($n = 80$), or by conventional insulin therapy ($n = 79$) with two daily subcutaneous insulin injections (clinical data are given in Table 1).

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Table 1. Clinical data and concentration of blood glucose, HbA_{1c}, immunoreactive insulin, C-peptide and circulating adhesion molecules of NIDDM patients and age-matched healthy subjects

	NIDDM patients	Healthy subjects
<i>n</i>	159	71
(Male, female)	(74/85)	(31/40)
Age (years)	62 ± 11	58 ± 7
Blood glucose level (mmol/l)	8.11 ± 3.06	4.89 ± 0.78 ^b
HbA _{1c} (%)	7.8 ± 1.5	5.5 ± 0.2 ^b
Serum insulin (pmol/l)	162 ± 62	96 ± 84 ^a
Serum C-peptide (nmol/l)	0.96 ± 0.69	1.06 ± 0.50
cICAM-1 (ng/ml)	315 ± 147	232 ± 58 ^c
cVCAM-1 (ng/ml)	886 ± 340	779 ± 224 ^a
cELAM-1 (ng/ml)	73 ± 36	43 ± 19 ^c

Values are means ± SD

^a $p < 0.05$; ^b $p < 0.01$; ^c $p < 0.001$ vs control subjects

Serum samples for determination of cICAM, cVCAM and cELAM-1 were analysed in duplicate by commercially available enzyme-linked immunosorbent assay (ELISA kits) for soluble ICAM-1, VCAM-1 and ELAM-1 following the supplied instruction manual (British Biotechnology Products Ltd, Abingdon, Oxon., UK), with both inter- and intra-assay coefficients of variation less than 9%. Two different serum pools were used as internal standards and an equal number of patients' and control subjects' sera were assayed on the same plate in order to minimize differences in interassay variations. Group differences were determined by Kruskal-Wallis or Wilcoxon 2-sample test. Correlations were estimated according to Pearson and Spearman. p -values less than 0.05 were considered to be significant.

Serum concentrations of cICAM-1, cVCAM-1 and cELAM-1 were elevated by 36, 14 and 70%, respectively, in NIDDM patients vs their control subjects (cICAM-1 (mean ± SD): 315 ± 147 vs 232 ± 58, $p < 0.001$; cVCAM-1: 886 ± 340 vs 779 ± 224, $p < 0.05$; cELAM-1: 73 ± 36 vs 43 ± 19 ng/ml, $p < 0.001$) (Table 1). Percentage of NIDDM patients exceeding the respective upper normal range (calculated as mean ± 2 SD of data in age matched control subjects) were 25% for cICAM-1, 11% for cVCAM-1 and 30% for cELAM-1. No difference in circulating adhesion molecules was apparent between NIDDM patients requiring insulin ($n = 79$) and those with no requirement ($n = 80$) (cICAM-1: 331 ± 166 vs 300 ± 125, NS; cVCAM-1: 925 ± 397 vs 847 ± 269, NS; cELAM-1: 71 ± 37 vs 75 ± 35 ng/ml, NS), nor in NIDDM patients with ($n = 70$) vs those without clinically symptomatic macroangiopathy ($n = 89$) as judged by resting ECG, a previous history of cardiac angina, intermittent claudication, myocardial or cerebral infarction (cVCAM-1: 900 ± 302 vs 889 ± 398, NS; cICAM-1: 300 ± 111 vs 344 ± 192, NS; cELAM-1: 72 ± 35 vs 77 ± 40 ng/ml, NS).

In agreement with results of Cominacini et al. [5] a weak correlation was seen between metabolic control as quantified by HbA_{1c} and cELAM-1 ($r = 0.19$; $p < 0.02$), but not cICAM-1 and cVCAM-1, in the group of NIDDM patients, while circulating adhesion molecules did not relate to concentrations of actual blood glucose, insulin and C-peptide. Moreover, circulating adhesion molecules were strongly related to each other in NIDDM patients (cVCAM-1 vs cICAM-1: $r = 0.73$, $p < 0.0001$; cICAM-1 vs cELAM-1: $r = 0.36$, $p < 0.0001$; cVCAM vs cELAM-1: $r = 0.30$, $p < 0.0001$), but less so in healthy subjects (cICAM-1 vs cELAM-1: $r = 0.36$,

$p < 0.01$; cVCAM-1 vs cICAM-1: NS; cVCAM-1 vs cELAM-1: NS).

Interestingly, cVCAM-1, but not cICAM-1 or cELAM-1, was significantly age-related both in NIDDM patients ($r = 0.26$, $p < 0.001$) and in healthy subjects ($r = 0.23$, $p < 0.05$) as previously described in non-diabetic patients with coronary and peripheral vascular disease [7].

Our findings of elevated concentrations of cICAM-1 (+36%), cVCAM-1 (+14%) and cELAM-1 (+70%) in NIDDM patients vs matched non-diabetic subjects support the hypothesis of a potential involvement of increased endothelial expression of adhesion molecules in macrovascular complications in diabetes mellitus and agree with reports of elevated cELAM-1 and cICAM-1 [5] and of elevated cELAM-1 and cVCAM-1 [6]. Since the degree of metabolic control as judged by HbA_{1c} levels is only weakly related to the observed elevation of circulating adhesion molecules in NIDDM patients, additional metabolic aberration such as increased oxidation of LDL-cholesterol or formation of advanced glycation endproducts may add to the appearance of elevated circulating adhesion molecules [8, 9]. The missing difference between concentrations of these molecules in patients with and without clinically symptomatic macroangiopathy may well be explained by the high incidence of clinically unnoticed atherosclerotic lesions in NIDDM [10].

In summary, we conclude that observed changes of circulating adhesion molecules in NIDDM patients may well reflect endothelial damage and leukocyte activation in a chronic diabetic milieu, and thus play a role in the pathogenesis of diabetic macroangiopathy.

Yours sincerely,

P. Fasching, W. Waldhäusl, O.F. Wagner

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Increased levels of plasma ELAM-1, ICAM-1 and VCAM-1 in NIDDM: possible role of oxidized LDL

Dear Sir,

Dr. Fasching et al. report very interesting data on circulating adhesion molecules in non-insulin-dependent diabetic (NIDDM) patients. Intercellular adhesion molecule-1 (ICAM-1), vascular cell adhesion molecule-1 (VCAM-1), and endothelial leukocyte adhesion molecule-1 (ELAM-1) were analysed in a large group of NIDDM patients ($n = 159$) and age-matched healthy subjects ($n = 71$). Circulating concentrations of ICAM-1, VCAM-1 and ELAM-1 were found to be elevated by 36, 14 and 70 % respectively in NIDDM patients vs the control subjects. Given the high number of subjects studied, the authors were also able to calculate that the percentage of NIDDM patients exceeding the respective normal range were 25 % for ICAM-1, 11 % for VCAM-1 and 30 % for ELAM-1. The results presented by Fasching et al. are in agreement with and strongly support the data previously reported by Steiner et al. [1] and Cominacini et al. [2].

In agreement with our results [2] and in disagreement with the paper of Steiner et al. [1], Dr. Fasching et al. found a weak correlation between ELAM-1 concentration and glycaemic control. They attributed the weakness of this correlation ($r = 0.19$, $p < 0.02$) to interfering metabolic abnormalities. Unfortunately, in this study, the authors speculate on the possible role of oxidized LDL and advanced glycation end products, but do not consider the simplest metabolic and non-metabolic abnormalities that could be the cause. Recent papers, for instance, demonstrated that hypertension [3] and dyslipidaemia [2] are pathological conditions which are also characterized by increased levels of plasma ELAM-1.

Dr. Fasching et al. considered increased oxidation of LDL or formation of advanced glycation end products to be the metabolic aberrations that could interfere with the correlation between ELAM-1 and glycaemic control.

As far as the increased oxidation of LDL is concerned, there is now data to demonstrate that the susceptibility of LDL to oxidation is increased in patients at risk of developing, or with established, atherosclerotic disease [4]. An increased susceptibility to LDL oxidation has also been shown in diabetic patients [5]. Even if 'susceptibility to LDL oxidation' is a semantic term that has never been clearly defined, and no general consensus exists as to how it should be determined, it may be related to the oxidation of LDL in the subendothelial space in vivo. Previous studies have shown that LDL tends to accumulate in areas of predilection in cholesterol-fed animals and that its residence time is increased [6]. This increase might allow for cellular oxidation of the LDL and the extent of this

process may be dependent on its susceptibility to oxidation. This view is supported by the presence in atheromatous lesions of LDL with characteristics similar to those of oxidatively modified LDL [7]. The mechanism by which oxidized LDL can accelerate the atherogenic process is unknown. Studies using cultured cells have demonstrated that oxidized LDL is chemotactic for monocytes and stimulates monocyte endothelial interactions [8]. Furthermore, Khan et al. [9] have recently demonstrated that modified LDL augments cytokine-activated VCAM-1 gene expression in human vascular endothelial cells. There may be a relation, therefore, between oxidized LDL and expression of some adhesion molecules. Future studies on the correlation between the susceptibility of LDL to oxidation and the concentration of soluble adhesion molecules in diabetic patients are required.

Yours sincerely,

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