Original Article

Serum Visfatin Increases With Progressive β-Cell Deterioration

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Visfatin has shown to be increased in type 2 diabetes but to be unrelated to insulin sensitivity. We hypothesized that visfatin is associated with insulin secretion in humans. To this aim, a cross-sectional study was conducted in 118 nondiabetic men and 64 (35 men and 29 women) type 2 diabetic patients. Type 1 diabetic patients with long-standing disease (n = 58; 31 men and 27 women) were also studied. In nondiabetic subjects, circulating visfatin (enzyme immunoassay) was independently associated with insulin secretion (acute insulin response to glucose [AIRg] from intravenous glucose tolerance tests) but not with insulin sensitivity (S_i) or other metabolic or anthropometric parameters, and AIRg alone explained 8% of visfatin variance (β = −0.29, P = 0.001). Circulating visfatin was increased in type 2 diabetes (mean 18 [95% CI 16–21] vs. 15 ng/ml [13–17] for type 2 diabetic and nondiabetic subjects, respectively; P = 0.017, adjusted for sex, age, and BMI), although this association was largely attenuated after accounting for HbA_{1c} (A1C). Finally, circulating visfatin was found to be increased in patients with long-standing type 1 diabetes, even after adjusting for A1C values (37 ng/ml [34–40]; P < 0.0001, adjusted for sex, age, BMI, and A1C compared with either type 2 diabetic or nondiabetic subjects). In summary, circulating visfatin is increased with progressive β-cell deterioration. The study of the regulation and role of visfatin in diabetes merits further consideration. *Diabetes* 55:2871–2875, 2006

Visfatin (also known as pre–B-cell colony-enhancing factor [1]) is a novel adipokine that is predominantly secreted by visceral adipose tissue (2), although controversy exists over the contribution of this fat depot to serum visfatin in humans (2,3). The protein exerts adipogenic effects in vitro and therefore is a good candidate to explain the accumulation of visceral adipose tissue that is associated with insulin resistance (2). Unexpectedly, insulin-mimetic effects were documented for this new adipokine, which are mediated by direct binding and activation of the insulin receptor (2).

In humans, plasma visfatin is increased in type 2 diabetes (4); however, studies to date (3–6) have failed to demonstrate an association of the circulating protein with insulin sensitivity. Because abnormalities in insulin secretion also contribute to the development of the metabolic abnormalities observed in type 2 diabetes, we hypothesized that besides insulin sensitivity, plasma visfatin levels are related to insulin secretion. Our results not only support previous reports of a lack of association of circulating visfatin with measures of insulin sensitivity but also show that β-cell dysfunction possibly mediates the link of this adipocytokine with diabetes.

RESEARCH DESIGN AND METHODS

One hundred and eighteen nondiabetic men, consecutively enrolled in a prospective study of cardiovascular risk factors in our health area, were included in the present study. None of these participants had evidence of metabolic disease other than nonmorbid obesity. Indeed, type 2 diabetes was ruled out by an oral glucose tolerance test (OGTT) according to criteria from the American Diabetes Association. Exclusion criteria for this group were *1*) BMI ≥40 kg/m^2 and *2*) concurrence of any systemic disease or medication use.

Sixty-four (35 men and 29 women) type 2 diabetic patients, defined according to the above-mentioned American Diabetes Association criteria and prospectively recruited from diabetes outpatient clinics at the Girona Hospital, were also studied. All type 2 diabetic patients had stable metabolic control in the previous 6 months. Pharmacological therapy for these subjects included insulin (34%), oral hypoglycemic agents (48%), statins (41%), fibrates (9%), blood pressure–lowering agents (48%), aspirin (27%), and allopurinol (5%). Exclusion criteria for this group were *1*) clinically significant hepatic, renal, neurologic, endocrinologic, or other systemic disease, including malignancy; *2*) current clinical evidence of hemochromatosis; and *3*) acute major cardiovascular event in the previous 6 months. Type 1 diabetic patients with long-standing disease (n = 58; 31 men and 27 women; mean duration of diabetes 16 years [range 6–46]) and daily insulin doses >0.7 units/kg were also included in the study. Three of these patients had renal disease, and 12 patients exhibited retinopathy.

All subjects were of Caucasian origin, reported that their body weight had been stable for at least 3 months before the study, and were free from intercurrent illnesses or chronic infections at time of the study. Informed written consent was obtained after the purpose, nature, and potential risks of the study were explained to the subjects. The experimental protocol was approved by the ethics committee of the Hospital of Girona.

Subjects were studied in the postabsorptive state. BMI was calculated as weight in kilograms divided by the square of height in meters. Subjects' waists were measured with a soft tape midway between the lowest rib and the iliac crest; hip circumference, when available, was measured at the widest part of the gluteal region; and waist-to-hip ratio was accordingly calculated. Blood pressure was measured in the supine position on the right arm after a 10-min rest. A standard sphygmomanometer of appropriate cuff size was used, and the first and fifth phases were recorded. Values used in the analysis are the average of three readings taken at 5-min intervals.

Insulin sensitivity (S_i), insulin secretion (acute insulin response to glucose [AIRg]), and glucose effectiveness (S_g) were measured by frequently sampled intravenous glucose tolerance tests (FSIGTs) in nondiabetic and newly diagnosed type 2 diabetic subjects. In brief, the experimental protocol for

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AIRg, acute insulin response to glucose; FSIGT, frequently sampled intravenous glucose tolerance test; OGTT, oral glucose tolerance test.

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FSIGT studies started between 8:00 and 8:30 A.M. after an overnight fast. A butterfly needle was inserted into an antecubital vein, and patency was maintained with a slow saline drip. Basal blood samples were drawn at −30, −10, and −5 min, after which glucose (300 mg/kg body wt) was injected over 1 min starting at time 0, and insulin (0.03 units/kg Actrapid; Novo Nordisk, Bagsværd, Denmark) was administered at time 20 min. Additional samples were obtained from a contralateral antecubital vein up to 180 min, as previously described (7).

HDL cholesterol was quantified after precipitation with polyethylene glycol at room temperature. Total serum triglycerides were measured through the reaction of glycerol-phosphate-oxidase and peroxidase. Serum glucose concentrations were measured in duplicate by the glucose oxidase method using a Beckman Glucose Analyzer II (Beckman Instruments, Brea, CA). The coefficient of variation (CV) was 1.9%. Serum insulin concentrations were measured in duplicate by a monoclonal immunoradiometric assay (IRMA; Medgenix Diagnostics, Fleunes, Belgium). Intra- and interassay CVs were <7%. Serum visfatin concentrations were measured by an enzyme immunoassay (Phoenix Peptides, Karlsruhe, Germany). Sensitivity of the method is 0.1 ng/ml. The intra- and interassay CVs were <6%. HbA_{1c} (A1C) was measured by high-performance liquid chromatography (Bio-Rad, Muenchen, Germany) and a Jokoh HS-10 autoanalyzer.

Statistical methods. Statistical analyses were performed using SPSS 12.0 software. Unless otherwise stated, descriptive results of continuous variables are expressed as means ± SD for Gaussian variables and as median (interquartile range) for non-Gaussian variables. Parameters that did not fulfill normal distribution were mathematically transformed to improve symmetry for subsequent analyses. The relation between variables was analyzed by simple correlation (Pearson's test) and multiple regression in a stepwise manner. Unpaired Student's *t* test was used to seek differences between men and women. One-way ANOVA followed by ANCOVA using general linear models (in order to correct for effect modifiers, such as sex, age, BMI, or A1C) were used to seek differences in plasma visfatin concentrations among groups. Levels of statistical significance were set at $P < 0.05$. For a given value of $P = 0.05$, the study had an 80% power to detect significant correlations (Pearson's coefficient of at least 0.3) between serum visfatin and metabolic parameters in bilateral tests. The study was also powered to detect a significant difference in circulating visfatin of at least 0.5 SD between nondiabetic and type 2 diabetic subjects.

RESULTS

Circulating visfatin is independently associated with insulin secretion, but not with insulin sensitivity, in nondiabetic men. Clinical and biochemical variables of the study subjects are summarized in Tables 1 and 2. Circulating visfatin was significantly associated with AIRg ($r = -0.27$, $P = 0.002$) in nondiabetic men (Fig. 1). Similarly, circulating visfatin was negatively correlated with 30-min OGTT insulin in these subjects ($r = -0.24$, $P = 0.005$). No significant correlations were found between visfatin and age, BMI, waist circumference, S_g, A1C, and 2-h OGTT plasma glucose in nondiabetic subjects. Serum visfatin was also significantly correlated with fasting insulin ($r = -0.20$, $P = 0.042$) and S_I ($r = 0.19$, $P = 0.043$) in nondiabetic men. However, on stepwise multiple regression analyses, AIRg was the only predictor of circulating visfatin, explaining 8% of its variance in nondiabetic men (Table 3).

TABLE 1
Clinical and laboratory variables in nondiabetic subjects

Characteristics	Nondiabetic men
n	118
Age (years)	51.0 ± 11.6
BMI (kg/m^2)	26.9 ± 3.6
Waist-to-hip ratio	0.93 ± 0.07
Systolic blood pressure (mmHg)	125 ± 15
Diastolic blood pressure (mmHg)	80 ± 10
HDL cholesterol (mg/dl)	53 ± 13
Triglycerides (mg/dl)	87 (65–130)
Fasting glucose (mg/dl)	96 ± 10
Fasting insulin (mIU/l)	8.0 (6.2–11.5)
S_I (min · mIU/l^{-1} · 10^{-4})	2.3 (1.5–3.5)
S_g (min^{-1})	0.019 ± 0.006
AIRg (min · mIU · l^{-1})	370 (194–532)
Visfatin (ng/ml)	15 (13–18)

Data are means ± SD for Gaussian variables and median (interquartile range) for non-Gaussian variables. S_I, S_g, and AIRg are from FSIGTs.

Circulating visfatin in type 2 diabetic subjects. As previously shown (4), serum visfatin was found to be increased in type 2 diabetes (mean 18 [95% CI 16–21] vs. 15 ng/ml [13–17] for type 2 diabetic and nondiabetic subjects, respectively; $P < 0.0001$ nonadjusted, $P = 0.017$ adjusted for sex, age, and BMI). In these subjects, A1C was highly correlated with circulating visfatin ($r = 0.49$, $P < 0.0001$; Fig. 2), and adjusting for A1C significantly attenuated the difference in serum visfatin between type 2 diabetic and nondiabetic subjects (17 [15–20] vs. 16 ng/ml [14–19], respectively; $P = 0.46$ adjusted for sex, age, BMI, and A1C).

To get further insight into the association of plasma visfatin with type 2 diabetes, subjects were divided into known ($n = 47$) and newly diagnosed (by OGTT) ($n = 17$) type 2 diabetic patients (Fig. 3). Known type 2 diabetic, in contrast to newly diagnosed type 2 diabetic, subjects have increased A1C values (6.9 ± 1.3 vs. 5.0 ± 0.9 vs. 4.9 ± 0.4% for known type 2 diabetes, new type 2 diabetes, and no diabetes, respectively; $P < 0.0001$).

By studying only known type 2 diabetic subjects, we documented persistently increased circulating visfatin in

TABLE 2
Clinical and laboratory variables in diabetic patients

Characteristics	Type 2 diabetic men	Type 2 diabetic women	*P*	Type 1 diabetic men	Type 1 diabetic women	*P*
n	35	29	—	31	27	—
Age (years)	54.0 ± 10.3	59.0 ± 10.7	NS	46.0 ± 12.9	38.0 ± 17.4	NS
BMI (kg/m^2)	30.3 ± 4.2	32.7 ± 5.4	0.003	25.2 ± 4.2	25.5 ± 5.0	NS
Systolic blood pressure (mmHg)	135 ± 19	134 ± 19	NS	129 ± 20	128 ± 17	NS
Diastolic blood pressure (mmHg)	80 ± 10	81 ± 8	NS	75 ± 8	73 ± 12	NS
HDL cholesterol (mg/dl)	48 ± 13	53 ± 16	NS	59 ± 15	77 ± 18	0.003
Triglycerides (mg/dl)	156 (108–246)	160 (104–205)	NS	101 (79–170)	53 (40–83)	<0.0001
Fasting glucose (mg/dl)	118 ± 39	137 ± 40	NS	173 ± 95	167 ± 75	NS
A1C (%)	5.6 ± 1.4	7.0 ± 1.3	0.002	6.9 ± 1.2	7.6 ± 0.8	0.08
Visfatin (ng/ml)	17 (13–21)	22 (18–25)	0.001	34 (29–55)	35 (25–47)	NS

Data are means ± SD for Gaussian variables and median (interquartile range) for non-Gaussian variables. *P* values shown on the right for each category are from unpaired *t* tests for comparisons between men and women. NS, not significant.

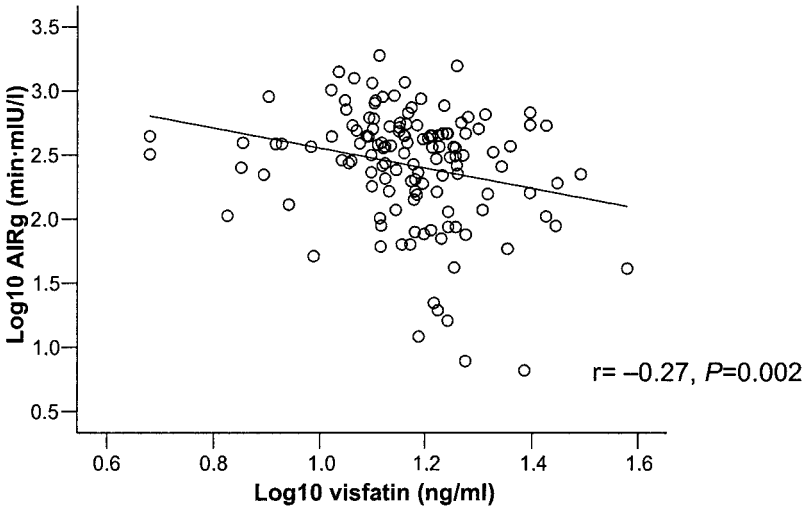

FIG. 1. Correlation graph of $\log_{10}$-transformed serum visfatin with $\log_{10}$-transformed AIRg (from FSIGT studies) in nondiabetic men. *r* and *P* values are shown from Pearson's analysis (nonadjusted data).

this metabolic disease, after accounting for the effect of the above-mentioned covariates (mean 19 [95% CI 16–22] vs. 15 ng/ml [13–17] for known type 2 diabetes and nondiabetic subjects, respectively; $P < 0.05$; Fig. 3). Newly diagnosed type 2 diabetes, on the other hand, exhibited circulating visfatin values that were similar to those of nondiabetic subjects (15 [10–19] vs. 15 ng/ml [13–17], respectively; P = NS, adjusted for sex, age, BMI, and A1C; Fig. 3). In accord with their A1C values, the latter patients showed only a partial defect in β-cell function, as evidenced by detectable AIRg values in most of these subjects (144 [0–343] vs. 444 [386–502] for new type 2 diabetic and nondiabetic subjects, respectively; $P = 0.004$ adjusted for sex, age, BMI, and A1C). On multiple regression analysis, A1C, but not sex, age, or BMI, was a predictor of circulating visfatin in type 2 diabetic patients, explaining 20% of its variance ($\beta = 0.457$, $P < 0.0001$).

Circulating visfatin is increased, but not associated with metabolic control, in type 1 diabetic subjects. To further test whether serum visfatin is dependent on preserved β-cell function, a group of type 1 diabetic patients with long-standing disease ($n = 58$; mean duration of diabetes 16 years [range 6–46]) were also studied (Table 2). Circulating visfatin was increased in type 1 diabetic vs. type 2 diabetic and nondiabetic subjects (37 [34–40] vs. 17 [15–20] and 16 ng/ml [14–19], respectively; $P < 0.0001$ adjusted for sex, age, BMI, and A1C compared with either type 2 diabetic or nondiabetic subjects; Fig. 3). Circulating visfatin, however, was unrelated to hyperglycemia (a potential confounding factor in the association between visfatin and β-cell function), as evidenced by a lack of correlation between serum visfatin concentration and A1C in type 1 diabetic subjects ($r = 0.17$, $P = 0.20$; Fig. 2*B*). On multiple regression analysis, age, but not sex, BMI, or A1C, was a predictor of circulating visfatin in type 1 diabetic patients, explaining 10% of its variance ($\beta = 0.344$, $P = 0.010$).

DISCUSSION

To our knowledge, this is the first report of an association between serum visfatin and β-cell dysfunction. Visfatin was recently shown to play a role in the amelioration of hyperglycemia in two different animal models of diabetes (2). Both overexpression of visfatin gene and treatment with recombinant protein exerted blood glucose–lowering effects in diabetic mice. Of note, recombinant visfatin was equipotent to exogenous insulin in this effect (2). To further support these findings, deletion of one of the two copies of the visfatin gene caused defects in glucose tolerance in mice (2). These observations, together with reports of increased serum visfatin in type 2 diabetes in both mice and humans (2,4), point to a potential relevant role of this novel adipocytokine in either the pathogenesis or the clinical phenotype of type 2 diabetes.

Recent efforts (3,4,8,9) to elucidate the role of visfatin in this disease have failed to describe an association of the adipocytokine with insulin sensitivity. Not surprisingly, visfatin was inconsistently related to anthropometrical and biochemical surrogates of insulin resistance, such as BMI, waist circumference, abdominal fat, fasting insulin, HOMA-IR, or serum adiponectin, in these and other studies (5,10). Similarly, genetic variations in the visfatin gene were only modestly associated with diabetes-related parameters, such as 2-h plasma glucose and fasting insulin concentration (11). Yet, the type 2 diabetes phenotype requires also a defect in β-cell function (12), which appears to has been overlooked in prior investigations.

Although insulin is unlikely to directly regulate visfatin serum concentrations (no effects on visfatin synthesis were documented after treating 3T3-L1 cells with insulin [13], no changes in plasma visfatin were observed upon fasting or feeding in mice [2], no differences in serum visfatin were seen in type 2 diabetic subjects treated with

TABLE 3
Multivariate regression analysis of serum visfatin as a dependent variable in nondiabetic men

Independent variables	Dependent variable $\log_{10}$ visfatin	
	β	*P*
Age	−0.068	0.432
BMI	0.036	0.678
Fasting insulin	−0.090	0.298
$\log_{10} S_I$	0.010	0.905
$\log_{10}$ AIRg	−0.287	0.001*

S_I and AIRg are from FSIGTs. *$R^2 = 0.076$.

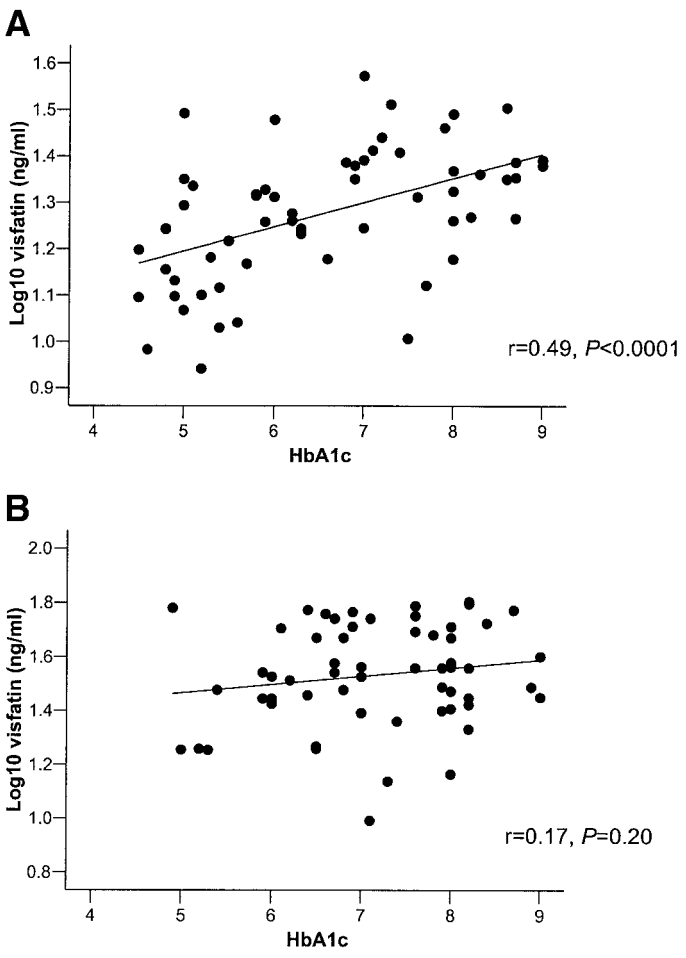

FIG. 2. Correlation graph of A1C with $\log_{10}$-transformed serum visfatin in type 2 (*A*) and in type 1 (*B*) diabetic patients. *r* and *P* values are shown from Pearson's analysis (nonadjusted data). Note: correlations were similar for male and female subjects; therefore, they are represented pooled in single groups.

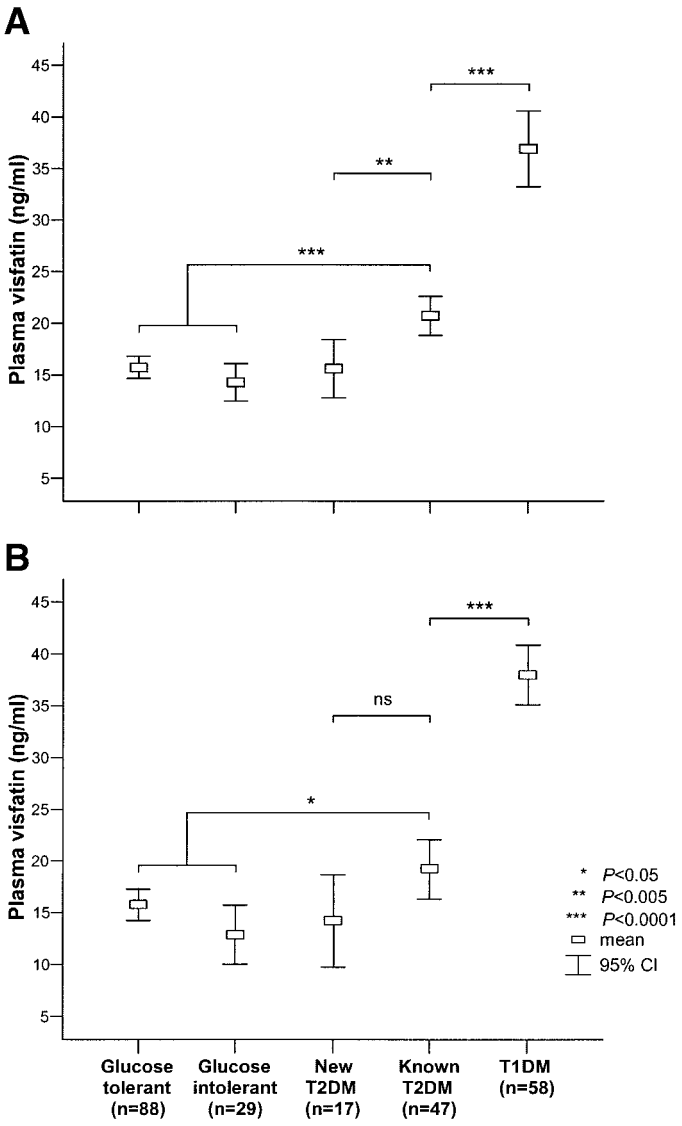

FIG. 3. Error-bar plots (means and 95% CIs) of serum visfatin in the different groups of subjects studied (data represent pooled male and female subjects for type 2 and type 1 diabetes). *A*: Nonadjusted data. *B*: Data adjusted for sex, age, BMI, and A1C.

sulfonylureas or exogenous insulin [4] and our own data [not shown], and no independent associations were observed between fasting insulin and serum visfatin in our study), our results are consistent with a physiopathological mechanism linked to β-cell dysfunction that mediates the association between increased circulating visfatin and both type 2 and type 1 diabetes. Besides its negative association with AIRg from FSIGT, serum visfatin was also negatively correlated with 30-min OGTT insulin (a surrogate index of insulin secretion) in our study. Furthermore, serum visfatin was recently shown to decrease following weight loss in morbidly obese subjects (10), an intervention that is believed to improve β-cell function (14,15). It should be noted that this mechanism appears not to be operative in the early phases of β-cell deterioration, as both glucose-intolerant and newly diagnosed type 2 diabetic subjects did not have increased serum visfatin in our study. A more severe defect, as seen in long-standing diabetes, is therefore needed to dysregulate circulating visfatin.

Circulating visfatin was also highly correlated with A1C in type 2 diabetic patients. Although factors related to hyperglycemia or oxidative stress can mediate in part the association between visfatin and diabetes (as indicated by attenuation of the difference in circulating visfatin between nondiabetic and type 2 diabetic subjects after adjusting for A1C), the fact that this difference persisted in known type 2 and in type 1 diabetic subjects, together with a lack of association between plasma visfatin and A1C in type 1 diabetic subjects, indicate that other factors besides hyperglycemia are involved in this relationship. On the other hand, increased A1C may reflect a more severe β-cell dysfunction (16), and, thus, adjusting for this factor will not surprisingly decrease the association of circulating visfatin with diabetes.

Among the counterregulatory hormones that are dysregulated in diabetic patients, both growth hormone and catecholamines were shown to be negative regulators of visfatin expression (13). Glucocorticoids, on the other hand, are known to induce visfatin secretion in adipocytes (13), and thus could contribute to the increased concentrations of this adipocytokine that accompany diabetes. It is also likely that other hormones of the entero-insular axis upregulate serum visfatin in the course of β-cell deterioration.

More recently, Haider et al. (17) have reported on the

effects on glucose and insulin on circulating visfatin in vivo and on visfatin secretion in vitro. Their results support our findings in type 2 diabetes by showing an upregulation of visfatin by hyperglycemia both in vivo and in vitro. Although insulin administration prevented the rise in visfatin concentrations under these conditions, insulin alone had no effect on either plasma visfatin or visfatin expression. Furthermore, other factors besides insulin must be involved in the regulation of plasma visfatin, as somatostatin, a general inhibitor of hormone secretion, was equipotent to insulin in preventing the rise in circulating visfatin following hyperglycemia in vivo (17).

As to the role of visfatin in human diabetes, insulin-mimetic actions are to be expected, as supported by the experimental animal data (2), and thus the increased serum visfatin concentration can respond to a compensatory mechanism aimed at ameliorating the functional consequences of insulin deficiency. The increased visfatin concentration may also promote insulin sensitivity by its stimulatory effects on peroxisome proliferator–activated receptor-γ and adiponectin gene expression (2). Yet, both unresponsiveness to visfatin actions (as seen for insulin and leptin) and a contribution of visfatin to β-cell deterioration cannot be ruled out in diabetic patients. In summary, circulating visfatin is increased with progressive β-cell deterioration. The study of the regulation and role of visfatin in diabetes merits further consideration.

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REFERENCES

1. Samal B, Sun Y, Stearns G, Xie C, Suggs S, McNiece I: Cloning and characterization of the cDNA encoding a novel human pre-B-cell colony-enhancing factor. *Mol Cell Biol* 14:1431–1437, 1994
2. Fukuhara A, Matsuda M, Nishizawa M, Segawa K, Tanaka M, Kishimoto K, Matsuki Y, Murakami M, Ichisaka T, Murakami H, Watanabe E, Takagi T, Akiyoshi M, Ohtsubo T, Kihara S, Yamashita S, Makishima M, Funahashi T, Yamanaka S, Hiramatsu R, Matsuzawa Y, Shimomura I: Visfatin: a protein secreted by visceral fat that mimics the effects of insulin. *Science* 307:426–430, 2005
3. Berndt J, Kloting N, Kralisch S, Kovacs P, Fasshauer M, Schon MR, Stumvoll M, Bluher M: Plasma visfatin concentrations and fat depot-specific mRNA expression in humans. *Diabetes* 54:2911–2916, 2005
4. Chen MP, Chung FM, Chang DM, Tsai JC, Huang HF, Shin SJ, Lee YJ: Elevated plasma level of visfatin/pre-B cell colony-enhancing factor in patients with type 2 diabetes mellitus. *J Clin Endocrinol Metab* 91:295–299, 2006
5. Hammarstedt A, Pihlajamaki J, Sopasakis VR, Gogg S, Jansson PA, Laakso M, Smith U: Visfatin is an adipokine but it is not regulated by thiazolidinediones. *J Clin Endocrinol Metab* 91:28–30, 2006
6. Arner P: Visfatin: a true or false trail to type 2 diabetes mellitus. *J Clin Endocrinol Metab* 91:28–30, 2006
7. Fernandez-Real JM, Broch M, Ricart W, Casamitjana R, Gutierrez C, Vendrell J, Richart C: Plasma levels of the soluble fraction of tumor necrosis factor receptor 2 and insulin resistance. *Diabetes* 47:1757–1762, 1998
8. Krzyzanowska K, Krugluger W, Mittermayer F, Rahman R, Haider D, Shnawa N, Schernthaner G: Increased visfatin concentrations in women with gestational diabetes mellitus. *Clin Sci (Lond)* 110:605–609, 2006
9. Pagano C, Pilon C, Olivieri M, Mason P, Fabris R, Serra R, Milan G, Rossato M, Federspil G, Vettor R: Reduced plasma visfatin/pre B-cell colony enhancing factor in obesity is not related to insulin resistance in humans. *J Clin Endocrinol Metab* 91:3165–3170, 2006
10. Haider DG, Schindler K, Schaller G, Prager G, Wolzt M, Ludvik B: Increased plasma visfatin concentrations in morbidly obese subjects are reduced after gastric banding. *J Clin Endocrinol Metab* 91:1578–1581, 2006
11. Bottcher Y, Teupser D, Enigk B, Berndt J, Kloting N, Schon MR, Thiery J, Bluher M, Stumvoll M, Kovacs P: Genetic variation in the visfatin gene (PBEF1) and its relation to glucose metabolism and fat depot specific mRNA expression in humans. *J Clin Endocrinol Metab* 91:2725–2731, 2006
12. DeFronzo RA: Lilly Lecture 1987: The triumvirate: β-cell, muscle, liver: a collusion responsible for NIDDM. *Diabetes* 37:667–687, 1988
13. Kralisch S, Klein J, Lossner U, Bluher M, Paschke R, Stumvoll M, Fasshauer M: Hormonal regulation of the novel adipocytokine visfatin in 3T3–L1 adipocytes. *J Endocrinol* 185:R1–R8, 2005
14. Guldstrand M, Ahren B, Adamson U: Improved beta-cell function after standardized weight reduction in severely obese subjects. *Am J Physiol Endocrinol Metab* 284:E557–E565, 2003
15. Camastra S, Manco M, Mari A, Baldi S, Gastaldelli A, Greco AV, Mingrone G, Ferrannini E: β-Cell function in morbidly obese subjects during free living: long-term effects of weight loss. *Diabetes* 54:2382–2389, 2005
16. Yki-Jarvinen H: Glucose toxicity. *Endocr Rev* 13:415–431, 1992
17. Haider DG, Schaller G, Kapiotis S, Maier C, Luger A, Wolzt M: The release of the adipocytokine visfatin is regulated by glucose and insulin. *Diabetologia* 49:1909–1914, 2006