Hyperglycemia modulates angiotensinogen gene expression

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Gabriely, Ilan, Xiao Man Yang, Jane A. Cases, Xiao Hui Ma, Luciano Rossetti, and Nir Barzilai. Hyperglycemia modulates angiotensinogen gene expression. **Am J Physiol Regulatory Integrative Comp Physiol** 281: R795–R802, 2001.—Elevated plasma angiotensinogen (AGT) levels have been demonstrated in insulin-resistant states such as obesity and type 2 diabetes mellitus (DM2), conditions that are directly correlated to hypertension. We examined whether hyperinsulinemia or hyperglycemia may modulate fat and liver AGT gene expression and whether obesity and insulin resistance are associated with abnormal AGT regulation. In addition, because the hexosamine biosynthetic pathway is considered to function as a biochemical sensor of intracellular nutrient availability, we hypothesized that activation of this pathway would acutely mediate in vivo the induction of AGT gene expression in fat and liver. We studied chronically catheterized lean (~300 g) and obese (~450 g) Sprague-Dawley rats in four clamp studies (n = 3/group), creating physiological hyperinsulinemia (~60 μU/ml, by an insulin clamp), hyperglycemia (~18 mM, by a pancreatic clamp using somatostatin to prevent endogenous insulin secretion), or euglycemia with glucosamine infusion (GlcN; 30 μmol·kg–1·min–1) and equivalent saline infusions (as a control). Although insulin infusion suppressed AGT gene expression in fat and liver of lean rats, the obese rats demonstrated resistance to this effect of insulin. In contrast, hyperglycemia at basal insulin levels activated AGT gene expression in fat and liver by approximately threefold in both lean and obese rats (P < 0.001). Finally, GlcN infusion simulated the effects of hyperglycemia on fat and liver AGT gene expression (2-fold increase, P < 0.001). Our results support the hypothesis that physiological nutrient "pulses" may acutely induce AGT gene expression in both adipose tissue and liver through the activation of the hexosamine biosynthetic pathway. Resistance to the suppressive effect of insulin on AGT expression in obese rats may potentiate the effect of nutrients on AGT gene expression. We propose that increased AGT gene expression and possibly its production may provide another link between obesity/insulin resistance and hypertension.

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# INSULIN RESISTANCE IS FREQUENTLY ASSOCIATED WITH OBESITY, HYPERTENSION, AND DYSLIPIDEMIA, WHICH REPRESENT MAJOR RISK FACTORS FOR ATHEROSCLEROTIC CARDIOVASCULAR DISEASE (25). THE RELATIONSHIP BETWEEN INSULIN RESISTANCE AND HYPERTENSION HAS BEEN DEMONSTRATED IN A VARIETY OF EPIDEMIOLOGICAL STUDIES (23, 34, 26). THE UNDERLYING MECHANISM, HOWEVER, IS FAR FROM BEING ELUCIDATED. IT HAS BEEN SPECULATED THAT HYPERINSULINEMIA INCREASES AGT EXPRESSION IN TISSUES OTHER THAN THE LIVER [WHICH IS ASSOCIATED WITH INSULIN RESISTANCE AND OBESITY], WHICH MIGHT MODULATE BLOOD PRESSURE THROUGH INCREASED VASCULAR SYPHOMATIC TONE OR BY REDUCING RENAL SODIUM EXCRETION (14). ANOTHER THEORY IS THAT DIRECT RENAL COMPRESSIVE TONE IN OBESITY INDUCES LOCAL ACTIVATION OF THE RENIN-ANGIOTENSIN SYSTEM (RAS) AND SUBSEQUENTLY HYPERTROPHY (17). RECENTLY, IT HAS BEEN DEMONSTRATED THAT HYPERLEPTINEMIA, WHICH IS COMMONLY ASSOCIATED WITH OBESITY, MAY INDUCE AN INCREASE IN THE SYMPATHETIC TONE AND MAY MODULATE BLOOD PRESSURE (19). HOWEVER, NONE OF THESE THEORIES HAS BEEN CLEARLY SHOWN TO HAVE A CAUSAL RELATIONSHIP.

The RAS plays a key role in the regulation of blood pressure (31). Angiotensinogen (AGT) is cleaved by renin and subsequently by angiotensin-converting enzyme (ACE) to angiotensin II, a potent vasoconstrictor and modulator of blood pressure (20). Overactivity of the AGT gene has been implicated in the development and maintenance of hypertension in rats (32, 37). In addition, it has been recently demonstrated that administration of AGT antisense mRNA to hypertensive rats induces a profound reduction in their blood pressure (33). Thus it appears that increased activity of the RAS may contribute to the development of hypertension. Although AGT is produced mainly in the liver, its genetic message is also abundant in brown and white adipose tissues (31). In fact, in human adipocytes, the gene expression of all components of the RAS has been demonstrated (8). The strong correlation between circulating AGT levels and body weight/insulin sensitivity suggests a central role for AGT in obesity and hypertension (7). However, in vitro studies have clearly demonstrated that insulin per se induces a downregulation of AGT gene expression in adipose cells, rejecting the hypothesis that hyperinsulinemia in obesity directly increases AGT (2, 1). In addition, AGT gene expression has been demonstrated to be regulated by nutritional stimuli. Earlier studies have shown that adipocyte AGT mRNA expression decreased after 3 days of fasting and was normalized after 6 days of fasting.
refeeding in rats (11), suggesting that nutrients may induce an upregulation of AGT.

The first aim in this study was to examine whether obesity and insulin resistance are associated also with resistance to the effect of insulin to suppress adipocyte AGT gene expression, a finding that may contribute to overexpression of adipocyte AGT. The second aim of the study was to examine whether adipocyte AGT gene expression may be acutely modulated in vivo by nutrients (glucose) and whether the hexosamine biosynthetic pathway (which is considered to function as a biochemical sensor of intracellular nutrient availability) may be implicated in adipocyte AGT gene regulation.

MATERIALS AND METHODS

Animals. Male Sprague-Dawley rats (Charles River Laboratories, Wilmington, MA) were housed in individual cages and were subjected to a standard light (6:00 AM to 6:00 PM)-dark (6:00 PM to 6:00 AM) cycle. All rats were fed ad libitum using regular rat chow that consisted of 64% carbohydrate, 30% protein, and 6% fat with a physiological fuel value of 3.3 kcal/g chow. Rats were studied during young adulthood before (~300 g body wt, 3 mo old, n = 12) and after (~450 g body wt, 5 mo old, n = 12) developing obesity (Table 1). We specifically used this model of rats because no dietary manipulation was needed to obtain obese and lean rats, and the difference in age between the two groups (7 and 13% of their maximal life span) was considered negligible in regard to the goals of the study. One week before the in vivo study, rats were anesthetized by inhalation of methoxyflurane, and indwelling catheters were inserted in the right internal jugular vein and in the left carotid artery (3, 6, 15, 18). This method of anesthesia allows fast recovery and normal food consumption after 1 day. The venous catheter extended to the level of the right atrium, and the arterial catheter was advanced to the level of the aortic arch. Recovery was continued until body weight was within 3% of the preoperative weight (~4–6 days). These chronically catheterized rats were studied after ~24 h of fasting while awake and unstressed.

Body composition was assessed as in Refs. 6, 15, and 16. Briefly, rats received an intra-arterial bolus injection of 20 μCi of tritiated-labeled water (3H2O; New England Nuclear, Boston, MA), and plasma samples were obtained at 30-min intervals for 3 h. Steady-state conditions for plasma 3H2O specific activity were achieved within 45 min in all studies. Five plasma samples obtained between 1 and 3 h were used in the calculation of the whole body distribution space of water.

Table 1. Body composition and basal metabolic characteristics of lean and obese Sprague-Dawley rats

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Lean</th>
<th>Obese</th>
</tr>
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<tbody>
<tr>
<td>Body wt, g</td>
<td>300 ± 10</td>
<td>465 ± 15*</td>
</tr>
<tr>
<td>LBM, g</td>
<td>276 ± 15</td>
<td>361 ± 16*</td>
</tr>
<tr>
<td>FM, g</td>
<td>24 ± 4</td>
<td>104 ± 7*</td>
</tr>
<tr>
<td>Liver, g</td>
<td>6 ± 1</td>
<td>8 ± 1*</td>
</tr>
<tr>
<td>Glucose, mM</td>
<td>6.8 ± 0.1</td>
<td>6.9 ± 0.1</td>
</tr>
<tr>
<td>Insulin, μU/ml</td>
<td>12 ± 3</td>
<td>27 ± 5*</td>
</tr>
</tbody>
</table>

All values expressed as means ± SE; n = 12 in each group. Body weight, lean body mass (LBM), fat mass (FM), liver mass, and plasma levels of glucose and insulin in lean and obese rats are shown. *P < 0.001 vs. lean rats.
carried out in a 50-mL reaction mixture containing 4 M. The expected RT-PCR product was 404 bp. PCR was 95°C, and the downstream primer was 5'-GCT TCT CCC AGC TGA CTG GG-3'. The sequence of the upstream primer for AGT was first-strand cDNA synthesis (GIBCO BRL) with random synthesized from 5'-2.2 M formaldehyde before use. The first-stranded cDNA was prepared from frozen tissues using TRIzol reagent (GIBCO BRL) as previously described (28). RNA from white adipose tissue was extracted from all rats (n = 3 in each group). Total hepatic RNA was prepared from frozen tissues using TRIzol reagent (GIBCO BRL) as previously described (28). RNA from white adipose tissue was prepared following Clontech’s protocol (18). The total RNA quality was analyzed by 1% agarose gel containing 2.2 M formaldehyde before use. The first-stranded cDNA was synthesized from 5 µg total RNA in 20-µl final incubation volume by using the SuperScript Preamplification System for first-strand cDNA synthesis (GIBCO BRL) with random primer. The sequence of the upstream primer for AGT was 5'-GCT TCT CCC AGC TGA CTG GG-3', and the downstream primer was 5'-GCT TGG TGT CAC CCA TCT TGC C-3'. The expected RT-PCR product was 404 bp. PCR was carried out in a 50-µl reaction mixture containing 4 µl of the above first-stranded cDNA, 5 µl of 10 × PCR buffer (Mg²⁺, Boehringer), 1 µl of the 10 mM dNTP mix, 4 pmol of each primer and 2.5 U Taq DNA polymerase (GIBCO). The conditions for PCR were 94°C for 45 s, 58°C for 45 s, 72°C for 90 s (20 cycles) using GeneAmp PCR system 9600 (Perkin-Elmer). Each assay was repeated for 10, 20, and 30 cycles to establish linearity. Each experiment was repeated three times for each individual animal. As a control we used β-actin gene expression (RT-PCR), described in detail elsewhere (6, 35). Briefly, the sequence of the upstream primer for β-actin was 5'-TGA GAC CTT CAA CAC CCC AGC C-3', and the sequence of the downstream primer was 5'-GAG TAC TTG GGC TCA GGA GGA G-3'. The conditions for PCR were 94°C for 45 s, 60°C for 45 s, and 72°C for 2 min (25 cycles). Quantification of AGT and signals were done by scanning densitometry, normalized for β-actin signal, which does not typically change with glucose, insulin, or GlcN, to correct for loading irregularities.

Analytic procedures. Plasma glucose was measured by the glucose oxidase method (Glucose Analyzer II, Beckman Instruments, Palo Alto, CA). Plasma insulin was measured by radioimmunooassay, using rat and porcine insulin standards. Plasma [³H]glucose radioactivity was measured in duplicates in the supernatants of Ba(OH)₂ and ZnSO₄ precipitates of plasma samples (20 µl) after evaporation to dryness to eliminate titrated water. To measure plasma 2-[¹⁴C]deoxyglucose, samples were deproteinized, and an aliquot of the supernatant was counted in a double channel beta-counter after addition of 500 µl of water and 5 ml of liquid scintillation mixture. To measure subcutaneous fat 2-[¹⁴C]deoxyglucose, frozen tissue samples were weighed and dissolved in 0.5 ml of 1 M NaOH kept in a shaking water bath at 60°C for 1 h. After neutralization with 0.5 ml of 1 M of HCl, two aliquots were taken. One was deproteinized with Ba(OH)₂ and ZnSO₄ and the other with 6% HClO₄. The HClO₄ supernatants contained both phosphorylated and unphosphorylated 2-deoxyglucose, whereas the Ba(OH)₂ and ZnSO₄ supernatants contained only the unphosphorylated form. The difference in disintegrations per minute between the two supernatants measures the fat content of 2-deoxyglucose phosphate, thus determining the rate of glucose uptake (28).

Statistical analysis. All values shown are expressed as means ± SE. Statistical analyses were performed using analysis of variance in multiple comparisons. Correlations were calculated using Pearson’s least squares method. A P value < 0.05 was considered to be statistically significant. All statistical analyses were performed using SPSS for Windows.

RESULTS

Biochemical and metabolic characteristics of lean and obese rats. Total body weight, lean body mass, and fat mass were significantly higher in the obese rats compared with the lean rats (50, 30, and 330% higher, respectively, P < 0.001 for all; Table 1). Fasting plasma insulin levels were 125% higher in the obese animals (P < 0.001), reflecting obesity-related insulin resistance.

AGT gene expression during saline infusion. When expression of subcutaneous fat AGT obtained from lean rats was used as standard (100 ± 11%), obese rats demonstrated higher AGT gene expression (116 ± 12% of lean; Fig. 2). AGT gene expression from liver was 121 ± 13% compared with adipose tissue of lean rats, and 126 ± 14% in liver of obese animals. Thus there was no significant difference between fat and liver AGT gene expression in the obese and lean animals.

Glucose fluxes and AGT gene expression during hyperinsulinemia. Physiological hyperinsulinemia (~60 µU/ml) induced an increase in total glucose uptake (TGU) by approximately twofold and adipose tissue glucose uptake (AGU) by approximately fivefold compared with saline control in lean animals (Table 2, Fig. 3). In contrast, a similar degree of hyperinsulinemia in obese rats resulted in TGU that was only ~1.4-fold and
AGU that was only approximately threefold increased compared with the saline control in the obese animals, demonstrating insulin resistance (Table 2). While hyperinsulinemia suppressed AGT gene expression in lean animals by 30%, it paradoxically increased AGT gene expression in obese animals (10%, P < 0.01, Fig. 3). This suggests that obese rats are resistant to the effect of insulin to suppress adipocyte AGT gene expression. AGT gene expression in the liver mirrored that of adipose tissue. Interestingly, although hepatic glucose production during hyperinsulinemia was 4.9 ± 1.1 mg·kg⁻¹·min⁻¹ in the lean rats, it was 8.1 ± 1.2 mg·kg⁻¹·min⁻¹ in obese rats, demonstrating specific insulin resistance in the liver of obese animals.

Glucose fluxes and AGT gene expression during hyperglycemia. Hyperglycemia (~17.7 mM) induced an increase in TGU and AGU to similar rates observed during the insulin studies in the lean and obese rats, while plasma insulin levels were maintained at basal levels using somatostatin infusion (Table 2, Fig. 4). Fat AGT gene expression significantly increased during hyperglycemia by approximately threefold in the lean and obese animals, and liver AGT gene expression was similarly elevated (Fig. 4).

Glucose fluxes and AGT gene expression during GlcN infusion. We hypothesize that if the hexosamine biosynthetic pathway is the activator of AGT gene expression during hyperglycemia, then GlcN infusion during a similar time course may mimic the effects of hyperglycemia (Table 2, Fig. 5). Indeed, although GlcN infusion had no effect on TGU or AGU compared with saline control, fat AGT gene expression significantly increased by approximately twofold in lean and obese animals (P < 0.001). As with the hyperglycemia studies, AGT gene expression in liver increased to a similar degree.

**Table 2. Biochemical characteristics and glucose fluxes during Ins, Glc, GlcN, and saline infusions in lean and obese rats**

<table>
<thead>
<tr>
<th></th>
<th>Ins</th>
<th>Glc</th>
<th>GlcN</th>
<th>Saline</th>
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<tbody>
<tr>
<td>Glucose, mM</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lean</td>
<td>6.9 ± 0.1*</td>
<td>17.6 ± 0.3*</td>
<td>6.8 ± 0.1*</td>
<td>7.0 ± 0.4</td>
</tr>
<tr>
<td>Obese</td>
<td>6.8 ± 0.1*</td>
<td>17.8 ± 0.1*</td>
<td>6.9 ± 0.1*</td>
<td>6.7 ± 0.5</td>
</tr>
<tr>
<td>Insulin, μU/ml</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lean</td>
<td>58 ± 4*</td>
<td>13 ± 4</td>
<td>13 ± 2</td>
<td>11 ± 3</td>
</tr>
<tr>
<td>Obese</td>
<td>62 ± 3*</td>
<td>22 ± 3‡</td>
<td>26 ± 3‡</td>
<td>29 ± 5‡</td>
</tr>
<tr>
<td>TGU, R</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Lean</td>
<td>25 ± 4†</td>
<td>28 ± 4†</td>
<td>13 ± 2</td>
<td>12 ± 3</td>
</tr>
<tr>
<td>Obese</td>
<td>18 ± 3†</td>
<td>20 ± 6†</td>
<td>13 ± 2</td>
<td>13 ± 2</td>
</tr>
<tr>
<td>AGU, R</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lean</td>
<td>20 ± 2†</td>
<td>23 ± 4†</td>
<td>3 ± 1</td>
<td>4 ± 1</td>
</tr>
<tr>
<td>Obese</td>
<td>12 ± 1‡</td>
<td>14 ± 3‡</td>
<td>3 ± 1</td>
<td>4 ± 1</td>
</tr>
</tbody>
</table>

All values expressed as means ± SE. Plasma glucose and insulin levels, total glucose uptake (TGU; determined by [3-3H]glucose in mg·kg⁻¹·min⁻¹), and adipose glucose uptake (AGU; determined by 2-deoxyglucose) in subcutaneous tissue expressed in [mg·kg tissue⁻¹·min⁻¹(R)], during the last 1 h of the clamp. Glc, hyperglycemia; Ins, hyperinsulinemia; GlcN, glucosamine. *P < 0.001 vs. others, †P < 0.001 vs. GlcN and saline, ‡P < 0.01 vs. lean.

**DISCUSSION**

This study demonstrates that AGT gene expression in fat and liver is markedly increased by a short exposure to glucose, possibly through the hexosamine biosynthetic pathway. Furthermore, this study suggests that in obesity, nutritional stimuli may induce an amplified effect on fat and liver AGT gene expression, due to resistance to the suppressive effect of insulin on AGT gene expression. Although plasma AGT levels and blood pressure were not measured (as will be discussed later) these findings provide a biological

Fig. 3. Hyperinsulinemia and AGT gene expression in lean and obese rats. Insulin infusion rate was 3 mU·kg⁻¹·min⁻¹; plasma glucose was clamped at euglycemia for 3 h. Baseline, saline infusion. A: fat; B: liver. *P < 0.01 vs. lean for AGT gene expression.
mechanism for the association of insulin-resistant states with hypertension.

It is well established that AGT and other components of the RAS are expressed in human fat, and their expression increases in association with obesity (7, 8). However, the role of hyperinsulinemia as a cause of increased AGT production has been questioned by the in vitro demonstration that insulin and its analogs act as negative regulators of AGT gene expression (1, 2). Our in vivo data support these findings; physiological hyperinsulinemia induced a significant decrease in fat and liver AGT gene expression in the lean animals.

However, there was no change in AGT gene expression during hyperinsulinemia in the obese rats, which suggests that obesity may be associated with resistance to the effect of insulin to suppress AGT gene expression. It is important to underline that the plasma insulin concentrations achieved during the studies were in the physiological postmeal range, and the total exposure time (3 h) parallels an average postmeal duration. Thus the hyperinsulinemic studies mimicked a typical periprandial state (in the absence of concomitant hyperglycemia), demonstrating no effect of hyperinsulinemia on fat and liver AGT gene expression in obesity.

Fig. 4. Hyperglycemia and AGT gene expression in lean and obese rats. Glucose and somatostatin (SRIF) were infused for 3 h to increase plasma glucose concentration to ~18 mM and to suppress endogenous insulin secretion. AGT gene expression is significantly ($P < 0.001$) increased from baseline (saline infusion) in lean and obese animals. A: fat; B: liver.

Fig. 5. GlcN infusion and AGT gene expression in lean and obese rats. GlcN was infused for 3 h with no changes in plasma insulin or glucose levels (compared to saline control). AGT gene expression is significantly ($P < 0.001$) increased compared with the baseline (saline infusion) in lean and obese animals. A: fat; B: liver.
We next examined whether hyperglycemia and glucose flux into adipose tissue induces AGT gene expression independent of the rise in plasma insulin levels. This was examined by increasing plasma glucose concentrations to match the rates of glucose uptake to the rates observed with hyperinsulinemia alone. Somatostatin infusion was used to suppress endogenous insulin hypersecretion during hyperglycemia. Under these conditions, fat and liver AGT gene expression were markedly increased in both lean and obese rats, suggesting that hyperglycemia per se represents a potent stimulus for AGT gene expression, with a similar sensitivity in lean and obese rats. Taken together, these results indicate that during hyperglycemia and hyperinsulinemia, fat or liver AGT gene expression is normally regulated positively by glucose and negatively by insulin. However, in obesity, there appears to be resistance to the negative regulatory effect of insulin on AGT gene expression, which results in an "unopposed" positive regulation by hyperglycemia.

Similar examples in which plasma insulin and nutrients have apparent opposite regulatory functions on gene expression have been demonstrated in the past. We previously showed that although the hepatic enzyme glucose-6-phosphatase (G6Pase) is negatively regulated by insulin (5), its expression increases by short exposure to both glucose and free fatty acids (4, 21). Increases in G6Pase gene expression by these nutrients during relative hypoinsulinemia (as in diabetes mellitus) may further impair hepatic glucose production.

The hexosamine biosynthetic pathway has a major role in diverting hexose phosphates and glutamine to glycolysis and synthesis of glycoproteins, mainly in the liver (Fig. 1). Because it accounts for only a small percentage (1–3%) of the glucose metabolized in myocytes and adipocytes, the hexosamine biosynthetic pathway is an ideal candidate as a cellular nutrient "sensor" responding to energy availability and mediating different metabolic effects. Recently, we demonstrated that leptin gene expression in fat and muscle tissues is increased with enhanced in vivo flux through this pathway (4, 27), providing feedback to the central nervous system regarding the nutritional status of the body, decreasing appetite, and directing free fatty acids to β-oxidation. Plasminogen activator inhibitor-1 (PAI-1) is another example of a peptide with its gene expression modulated by induction of the hexosamine biosynthetic pathway. Similar induction of AGT gene expression by glucose and GlcN supports the role of this pathway in its induction (30). Recent evidence attributes a key role of SP-1 transcription factor in the regulation of gene expression by nutrients (12, 22, 27).

Although basal expression of AGT gene expression in fat and liver were expected to be increased with obesity, our experimental model yielded similar values to controls (Fig. 2). This may be explained by the short duration of obesity in our rat model (2 mo difference between the obese and lean rats) and possibly by the assumption that a relative increase in AGT gene expression in pulses (with meals) may induce a constant increase in plasma AGT levels, as demonstrated in various studies in humans (7, 8). We did not anticipate significant changes in AGT plasma levels or blood pressure after only 3 h of physiological induction by nutrients. AGT production (and plasma levels) may increase with repeated meals, resulting in an overall increase in AGT levels with obesity. The purpose of this study, however, was to examine mechanisms responsible for modulating fat and liver AGT gene expression, rather than the AGT protein.

Our data also support the notion that in obesity and diabetes mellitus there is a defect in physiological evening and nighttime blood pressure variations (10, 36), i.e., in these conditions, blood pressure fails to decrease during the night. Repeated stimulation of AGT by hyperglycemia (with meals) during the day may parallel with the time course needed for nutrients to activate AGT gene expression and, subsequently, secretion (24). Another example of a peptide with its gene expression regulated by nutrients and its plasma levels peaking at night is leptin, which also appears to be regulated by the hexosamine biosynthetic pathway (35). Indeed, leptin is an example of a fat-derived peptide whose blood levels increase gradually throughout the day and peak at night (36), an effect that is contributed to meals timing (29, 30). The hexosamine biosynthetic pathway may be responsible for ~50% increases in plasma leptin levels observed in humans throughout the day. Thus increases in the gene expression of these peptides during the day may result in an increase in their plasma concentration during the night, and consequently may induce the target clinical effects.

As far as we know, this is the first study to demonstrate an induction of fat and liver AGT gene expression by glucose, an effect that was mimicked by GlcN infusion. Similarly, our data demonstrate that obesity and insulin resistance are associated with resistance to the effect of insulin to suppress fat and liver AGT gene expression, findings that support previous studies that showed a direct correlation between obesity/diabetes mellitus, increased AGT plasma levels, and hypertension. Thus we suggest that the increased prevalence of hypertension in obesity and/or diabetes mellitus may be linked to this phenomenon.

**Perspectives**

Fat tissue seems to play a central role as a regulatory and secretory organ responsible for a variety of metabolic processes. Indeed an explosion of evidence has demonstrated that fat tissue is a large, very active endocrine gland, which secretes a variety of peptides [such as leptin, adipocyte complement-related protein (Acrp30), resistin, and PAI-1], cytokines (such as tumor necrosis factor), and complement factors (such as C3, and B). Although the expression of many of these peptides may be due to the fact that fat is derived from the same stem cells as bone marrow, we propose that many of these proteins may have a role in diseases when the following conditions are met: first, when fat...
mass is increased, as seen in >50% of the adult population in the U.S. where fat accounts for >50% of the body weight, and second, by nutrients through some specific sensing mechanisms as described here and previously (27). Such nutrient sensing pathway may be chronically activated by hyperglycemia in the diabetic patients, amplifying the risks for related diseases.

The epidemiological data in human obesity and diabetes mellitus are based on measuring fasting levels of substrates and proteins. Because nutritional regulation may act on the transcriptional level, plasma peptide concentrations will probably increase throughout the day and be maximal at night, as is the case for leptin. Similar to the determination of insulin sensitivity, which uses stimulation to outline insulin resistance, plasma peptide levels after feeding may be used to determine the relationship between elevated plasma peptides and the clinical consequences (e.g., AGT and hypertension, PAI-1, and coronary events). Thus we suggest that fat-derived peptides are a “first line” candidate to explore the relationship between obesity/diabetes and related diseases.

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REFERENCES


