Insulin Resistance and Body Fat Distribution

Contribution of visceral fat accumulation to the development of insulin resistance and atherosclerosis

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Body fat distribution can be assessed by computed tomography (CT). The ratio of visceral fat area to subcutaneous fat area (V/S ratio) at the level of umbilicus was used to classify obese subjects as having visceral fat obesity (VFO) or subcutaneous fat obesity (SFO). Serum triglyceride and total cholesterol levels and plasma glucose area in an oral glucose tolerance test were higher in patients with VFO than in those with SFO. Significant positive correlations were demonstrated between V/S ratio and plasma glucose area, serum triglyceride level, and total cholesterol level as well as systolic or diastolic blood pressure. VFO was more frequently associated with coronary artery disease. Moreover, VFO was more often accompanied by multiple risk factors than was SFO. Steady-state plasma glucose (SSPG) level was significantly higher in patients with VFO than with SFO, suggesting that insulin resistance may be more remarkable in VFO than in SFO.

Furthermore, visceral fat accumulation was also associated with these complications even in nonobese subjects. Visceral fat area (VFA) was significantly correlated with fasting plasma glucose, serum triglyceride, and total cholesterol levels. Animal models such as Goto-Kakizaki (GK) rats with ventromedial hypothalamus (VMH) lesions and Otsuka-Long-Evans-Tokushima-Fatty (OLETF) rats were accompanied by visceral fat accumulation and an early stage of aortic atherosclerosis. Aging, sex hormone, genetic, and dietary factors and physical inactivity may induce visceral fat accumulation. Visceral fat is characterized by its high lipogenic activity as well as its accelerated lipolytic activity. High levels of portal free fatty acids (FFAs) may eventually result in an enhancement of hepatic triglyceride synthesis, causing hyperlipidemia. High portal FFA levels would also induce insulin resistance, thereby causing glucose intolerance, hypertension, and finally atherosclerosis. We propose a term, "visceral fat syndrome," as a highly atherogenic state, which includes visceral fat accumulation, glucose intolerance (insulin resistance), hyperlipidemia, and hypertension.

In 1947, Vague (1) noted that the incidence of metabolic complications among equally obese subjects may vary depending on their physique. Morbidity was shown to be higher in android-type obesity than in gynoid-type obesity. Later, Kissebah et al. (2) proposed a classification of obesity, namely upper body segment obesity and lower body segment obesity, using waist-to-hip ratio, and demonstrated that upper body segment obesity is accompanied by metabolic complications such as diabetes and hyperlipidemia. The morbidity was higher in upper body segment obesity than in lower body segment obesity. In 1983 we established a method to measure body fat volume and fat distribution in obese subjects (3). We used a computed tomography (CT) method that permits an accurate analysis of subcutaneous fat and intra-abdominal visceral fat. Based upon this method, we demonstrated that patients with fat deposition in the abdominal cavity have a higher incidence of metabolic and vascular complications associated with obesity and proposed a novel classification of obesity: visceral fat obesity (VFO) and subcutaneous fat obesity (SFO) (4). Visceral fat accumulation was shown to be associated with insulin resistance. The idea of visceral fat accumulation was further extended to the pathogenesis of metabolic and cardiovascular complications in nonobese subjects. In the current report, we describe the relationship of visceral fat accumulation to various complications as well as the mechanism for deposition of visceral fat.

RESULTS AND DISCUSSION

Relation between VFO and metabolic and vascular complications

We suggested that the ratio of visceral fat area to subcutaneous fat area (V/S ratio) obtained from CT cross-sectional pictures of the umbilical region was the most useful determinant of complications associated with obesity. From the distribution of V/S ratio, we designated the subjects with a V/S ratio ≥0.4 as having VFO. The subjects were divided into two groups by considering V/S ratio, and these two groups were matched for age, sex, and BMI. Serum triglyceride and total cholesterol levels and plasma glucose area in an

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ACS, acyl-CoA synthetase; CAD, coronary artery disease; CT, computed tomography; FFA, free fatty acid; GK, Goto-Kakizaki; LPL, lipoprotein lipase; OLETF, Otsuka-Long-Evans-Tokushima-Fatty; SFA, subcutaneous fat area; SFO, subcutaneous fat obesity; SMC, smooth muscle cell; SSPG, steady-state plasma glucose; VFA, visceral fat area; VFO, visceral fat obesity; VMH, ventromedial hypothalamus; V/S ratio, ratio of visceral fat area to subcutaneous fat area.

Table 1—*Clinical and metabolic profiles of obese patients with CAD*

	Patient								
	1	2	3	4	5	6	7	8	9
Age (years)	49	59	55	37	51	42	67	69	63
BMI	33	31	30	29	28	31	29	27	28
Coronary angiographic findings	TVD	DVD	DVD	TVD	SVD	SVD	TVD	SVD	TVD
V/S ratio	1.1	1.25	1.74	0.42	0.96	0.81	1.21	1.79	1.19
Hypertension	+	−	+	−	+	+	+	+	−
Glucose intolerance	IGT	IGT	Diabetes	Normal	IGT	IGT	Normal	Diabetes	IGT
Hyperlipidemia	+	+	+	−	−	+	+	+	+

TVD, triple vessel disease; DVD, double vessel disease; SVD, single vessel disease; IGT, impaired glucose tolerance.

oral glucose tolerance test were all higher in patients with VFO than in those with SFO in both sexes (4). Significant positive correlations were demonstrated between V/S ratio and plasma glucose area, serum triglyceride level, and total cholesterol level (5).

In addition to these metabolic disorders, we demonstrated that visceral fat accumulation was also closely correlated with blood pressure (6) and left ventricular enlargement (7). Furthermore, when the number of accompanying coronary risk factors was compared between patients with these two types of obesity, VFO was more frequently accompanied by multiple risk factors than was SFO.

To test for the presence or absence of insulin resistance in VFO, steady-state plasma glucose (SSPG) levels were measured and compared between patients with VFO and SFO. SSPG level was significantly higher in those with VFO than in those with SFO (309 ± 49 mg/dl [mean ± SD] in VFO vs. 260 ± 40 mg/dl in SFO, $P < 0.05$), while no significant difference was seen in steady-state plasma insulin level between the two types of obesity. These data suggested that insulin resistance may be more remarkable in VFO than in SFO.

The contribution of visceral fat accumulation to the occurrence of coronary artery disease (CAD) was further investigated in 38 male patients with CAD for whom coronary angiographic examination and analysis of body fat distribution by CT scan were performed. Nine of 38 patients with CAD were obese, while the other 29 patients were not. The profiles of nine obese patients with CAD are shown in Table 1. When the V/S ratio in obese male and female patients was plotted with regard to the presence or absence of CAD, V/S ratios for most of the obese patients with CAD were extremely high when compared with those of obese subjects without CAD. CAD in many of the obese patients was accompanied by hypertension, impaired glucose tolerance or diabetes, and hyperlipidemia.

From these observations, we proposed and established a term, "visceral fat obesity," as a highly atherogenic state, which includes obesity, visceral fat accumulation, glucose intolerance, hyperlipidemia, and hypertension. Similar ideas of a multiple risk factor clustering syndrome have recently been advocated. The concept of VFO may overlap with that of syndrome X proposed by Reaven (8) or deadly quartet proposed by Kaplan (9). Patients with both of these syndromes have recently been noted as being susceptible to ischemic heart disease with a clustering of multiple risk factors.

Contribution of visceral fat accumulation to complications in nonobese subjects

We recognized that visceral fat area (VFA) was often increased even in nonobese subjects. The correlations of age, BMI, and fat area with metabolic profiles were assessed in nonobese subjects. Only VFA was significantly correlated with fasting plasma glucose, serum triglyceride, and total cholesterol levels, while the V/S ratio was not a good index of metabolic complications in nonobese subjects. When the correlations between VFA and systolic or diastolic blood pressure were assessed in subjects with normal body weight, good positive correlations were demonstrated between VFA and systolic or diastolic blood pressure. Interestingly, even in nonobese subjects, a substantial number of patients with CAD had a more marked accumulation of visceral fat than did control subjects without CAD. When VFA and subcutaneous fat area (SFA) were compared between nonobese subjects with and without CAD, there was no significant difference in SFA between nonobese CAD patients and nonobese control subjects. However, the mean VFA was significantly higher in nonobese CAD patients than in nonobese control subjects, and ~40% of nonobese patients with CAD had a remarkable increase in visceral fat. When the relationship between visceral fat accumulation and the number of diseased coronary arteries was investigated, VFA was increased in proportion to the number of diseased vessels. A positive correlation was noted between VFA and coronary extent score, which indicates the severity of coronary atherosclerosis. Among nonobese patients with CAD, high VFA was more frequently accompanied by multiple risk factors than was normal VFA (Table 2), which was similar to the results obtained from obese patients with CAD.

With regard to glucose metabolism in nonobese subjects, plasma glucose and insulin areas were compared between nonobese subjects with and without CAD. As shown in Fig. 1, both plasma glucose area and plasma insulin area were significantly higher in nonobese patients with CAD than in nonobese control subjects, suggesting the presence of insulin resistance in nonobese subjects with CAD. Therefore, we propose the term "visceral fat syndrome," which includes visceral fat accumulation, glucose intolerance, hyperlipidemia, and hypertension, as a highly atherogenic state with a cluster of risk factors based upon visceral fat accumulation, irrespective of body weight. This syndrome might be likened to a wa-

Table 2—*Metabolic characteristics, blood pressure, and the number of risk factors in nonobese male subjects with normal and high VFA*

	Normal VFA (VFA <100 cm^2)	High VFA (VFA ≥100 cm^2)
n	79	67
Total cholesterol (mg/dl)	175 ± 35	202 ± 45*
Triglycerides (mg/dl)	127 ± 72	152 ± 99
HDL cholesterol (mg/dl)	52 ± 17	49 ± 14
Fasting plasma glucose (mg/dl)	107 ± 38	114 ± 36
Systolic blood pressure (mmHg)	126 ± 18	138 ± 17*
Diastolic blood pressure (mmHg)	77 ± 12	85 ± 13*
Risk factor		
None	41 (52)	9 (13)
Single	21 (27)	24 (36)
Multiple	17 (21)	34 (51)†

Data are means ± SD or *n* (%). Risk factor: hyperlipidemia (total cholesterol ≥220 mg/dl and/or triglycerides ≥150 mg/dl); hyperglycemia (fasting plasma glucose ≥110 mg/dl); hypertension (systolic blood pressure ≥140 and/or diastolic blood pressure ≥90 mmHg). *$P < 0.001$. †$P < 0.01$ (3 × 2 contingency table).

terfall, such as Niagara Falls. From the upper stream of visceral fat accumulation, dependent on or independent from insulin resistance, hypertension, glucose intolerance, or dyslipidemia cascade down, finally developing atherosclerosis as a basis of a waterfall.

Animal models for visceral fat accumulation

To further elucidate the mechanism for the development of atherosclerosis related to visceral fat accumulation, it may be necessary to develop appropriate animal models for visceral fat syndrome. So far, two models have been investigated: one is GK rats with ventromedial hypothalamus (VMH) lesions, and the other is OLETF rats. The GK rat has already been widely used as a model of insulin resistance. Using this model, a VMH lesion was developed to induce hyperphagia (10). The intake of food was greatly increased 1–2 weeks after the VMH operation, and then it was gradually reduced. However, the rate of body weight gain was significantly lower in the GK rat with a VMH lesion than in the GK rat with a sham operation. The VMH lesion in GK rats caused a marked deterioration of glucose metabolism. The plasma glucose level began to increase 1 week after the operation, reaching a plateau at 2 weeks. The VMH lesion caused an enhancement of insulin reaction at the early stage, but thereafter insulin secretion was reduced with the depletion of pancreatic insulin content. Hypertriglyceridemia was also noted in the GK rats with VMH lesions.

When fat distribution was compared between GK rats with VMH lesions and sham operation, the mesenteric fat weight was significantly increased in GK rats with VMH operations compared with GK rats with sham operation (9.0 ± 2.0 g in GK rats with VMH operation vs. 7.2 ± 1.0 g in GK rats with sham operation, respectively; $P < 0.05$). In contrast, no significant difference was noted in subcutaneous fat weight between the two groups. Therefore, this animal could be a model of visceral fat syndrome, although it is not complete since it is not accompanied by hypertension.

In the aorta of VMH-lesioned GK rats, a marked thickening of the intima with an infiltration of smooth muscle cells (SMCs) was observed (11). The migration of SMCs and an infiltration of lymphocytes into the intima were also demonstrated by electron microscopy. The thickness of the intima of the descending aorta was fourfold increased in the VMH-lesioned GK rats than in the GK rats with sham operations. The number of cells infiltrating into the intima of descending aorta in VMH-lesioned GK rats was also twice that in GK rats with sham operations. To elucidate the mechanism for the development of atherosclerosis in VMH-lesioned GK rats, the outgrowth rate of SMCs from the aorta of these rats was determined. It was markedly increased in GK rats with VMH lesions compared with that of sham-operated GK rats as well as that of control rats.

Another candidate for the animal model of visceral fat syndrome might be OLETF rats (12). This rat model is characterized by a late onset of hyperglycemia with insulin resistance, mild obesity, and hypertriglyceridemia. In this model, the accumulation of mesenteric fat was more prominent than that of subcutaneous fat when compared with accumulations in control rats named LETO. Slight intimal thickening and increased infiltration of cells were demonstrated in OLETF rats. Since streptozotocin-induced diabetic rats, a severe IDDM model, do not manifest such vascular changes, the early stage of atherosclerotic changes observed in these two models might be due to the metabolic disturbances related to visceral fat accumulation. We are currently investigating the mechanisms for the vascular changes in these animals.

Metabolic characteristics of visceral fat tissues

What are the characteristics of visceral fat tissues? Regarding lipogenesis, we investigated the changes in the expression of mRNA of lipoprotein lipase (LPL) and glucose transporter (GLUT4), which have important roles in energy influx to muscles and adipose tissues. We also investigated changes in the mRNA expression of acyl-CoA synthetase (ACS), which is considered to be an important enzyme for triglyceride formation in adipose tissues and for β-oxidation in muscles. After VMH operations were performed in Sprague-Dawley rats, the time course of changes in body weight and cell volume of fat tissues was analyzed (13). The mesenteric fat weight showed a trend for increasing on the 1st day after VMH operation, and the cell volume represented an ~1.4-fold in-

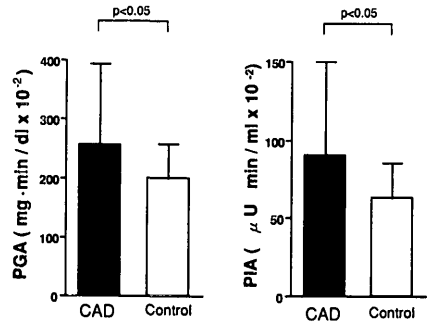

Figure 1—*Comparison of plasma glucose and insulin areas obtained by oral glucose tolerance tests between nonobese patients with CAD and control subjects. PGA, plasma glucose area; PIA, plasma insulin area.*

crease compared with that of control rats. There was no significant difference in fat weight and cell volume in subcutaneous fat at this stage. However, the rate of increase in tissue weight was much larger in mesenteric fat than in subcutaneous fat at 1 and 5 days after operation. The mesenteric fat represented an approximately fivefold increase in ACS activity 1 day after VMH operation, while no appreciable increase in ACS activity was noted in subcutaneous fat at this stage. The ACS activity in subcutaneous fat showed a slower response to VMH operation compared with that in visceral fat. Slot blot analysis showed that the expressions of both ACS and LPL mRNA were increased after VMH operation. When the increases in these mRNAs were compared between mesenteric fat and subcutaneous fat, mesenteric fat showed a more rapid increase in the expressions of ACS and LPL mRNA levels than in subcutaneous fat. These data indicate that the enhancement of ACS and LPL gene expression is important for fat accumulation at a very early stage and that intra-abdominal visceral fat tissues respond more rapidly than subcutaneous fat to VMH operation, leading to the accumulation of fat.

With regard to lipolytic activities of visceral fat tissues, mesenteric fat was shown to have a higher lipolytic activity than subcutaneous fat, when glycerol was liberated by norepinephrine (14). Furthermore, a good positive correlation was demonstrated between the relative weight of mesenteric fat and portal FFA level, suggesting that visceral fat may be metabolically more active than subcutaneous fat, releasing FFAs into the portal circulation.

Factors affecting visceral fat accumulation

In Japan, professional sumo wrestling continues to maintain its popularity as a national sport. Sumo wrestlers eat a very high-energy diet, comsuming up to 7,000 kcal/day to gain body weight; however, at the same time, they perform very hard physical exercise every day. Fat distribution and laboratory tests were analyzed in young sumo wrestlers. Interestingly, sumo wrestlers showed an accumulation of fat only in the subcutaneous area with a markedly developed musculature. However, visceral fat accumulation was very mild compared with that in obese male subjects with a similar BMI. The average V/S ratio of young sumo wrestlers was 0.25, which was comparable to that of patients classified as having SFO. Surprisingly, most of the sumo wrestlers maintained normal glucose and triglyceride levels and had unexpectedly low cholesterol levels despite marked obesity. These data suggested that physical exercise might have prevented visceral fat accumulation.

To elucidate the mechanism for the preventive effect of physical exercise, the influence of exercise on visceral fat accumulation was assessed in male Wistar rats performing 1 h of physical exercise on a treadmill for 7 days (15). Treadmill exercise training caused a significant reduction in mesenteric fat weight as well as in mesenteric adipocyte volume, while subcutaneous fat weight and adipocyte volume did not change significantly after exercise. On the contrary, muscle weight was significantly increased by exercise. ACS activity of the mesenteric fat was significantly lower in the exercised group than in the sedentary group. In contrast, ACS activity of subcutaneous fat showed no significant difference between the sedentary and exercised states. Regarding muscle tissues, ACS activity from the gastrocnemius muscle was higher in the exercised group than in the sedentary group. The increased ACS activity in gastrocnemius muscle was supposed to indicate a greater supply of acyl-CoAs for β-oxidation, through which the energy for muscle action is produced. Furthermore, Northern blot analysis of LPL, GLUT4, and ACS mRNA demonstrated that physical exercise induced a marked reduction in LPL, GLUT4, and ACS mRNA in mesenteric fat, while little change in these parameters was observed in subcutaneous fat and muscles. The data clearly suggested that physical exercise may switch the direction of energy flux from mesenteric fat tissues to muscles, depressing lipogenesis in mesenteric fat. This may be one of the mechanisms for the preventive effect of physical exercise against visceral fat accumulation.

CONCLUSIONS — Several factors, including sex hormones, aging, and genetic and dietary factors and especially a lack of exercise may induce visceral fat accumulation. Visceral fat is characterized by its high lipogenic activity as well as its increased lipolytic activity. High levels of portal FFAs may eventually result in an enhancement of hepatic triglyceride synthesis, causing hyperlipidemia. High portal FFA levels would also induce insulin resistance, thereby causing glucose intolerance, hypertension, and finally atherosclerosis. The term "visceral fat syndrome" may be more appropriate, since it clearly defines the cause of multiple risk factor clustering syndrome. Future studies may be necessary to elucidate the basis for the link between visceral fat accumulation and cardiovascular complications as well as to identify the genes that are specific for visceral adipose tissues.

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