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Different functions of visceral and subcutaneous fat cells

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Visceral fat accumulation is known to be an evident clinical index for the insulin resistance related with obesity. Patients with excessive accumulation of visceral fat frequently suffered from metabolic disorder, such as hyperlipidemia, hypertension, and glucose intolerance. However, molecular mechanism for the pathogenesis of obesity-accompanied metabolic disorders has not been fully elucidated. It has been clarified that adipocytes in visceral fat area have different functions from subcutaneous fat area, and these differences might contribute the pathological significance of excessive accumulation of visceral fat for the accompanied insulin resistance and hyperinsulinemia.

We established the unique method to clarify whether the functional differences between visceral and subcutaneous adipocytes depend on their anatomical location. 3T3-L1 cells or TNF-α overexpressing CHO cells were implanted into subcutaneous fat area or mesenteric area as visceral fat area in athymic mice of BALB/C strain (Figure 1). The serum concentrations of TNF-α increased gradually after implantation with 3T3-L1 cells into mesenteric area, and these reached to 141.6 ± 30.9 pg/ml at four weeks. However, the mice injected into subcutaneous area did not show any difference from those of Sham-operated mice. Body weight during experimental periods did not differ among the mice implanted with 3T3-L1 cells into mesenteric and subcutaneous area, and control.

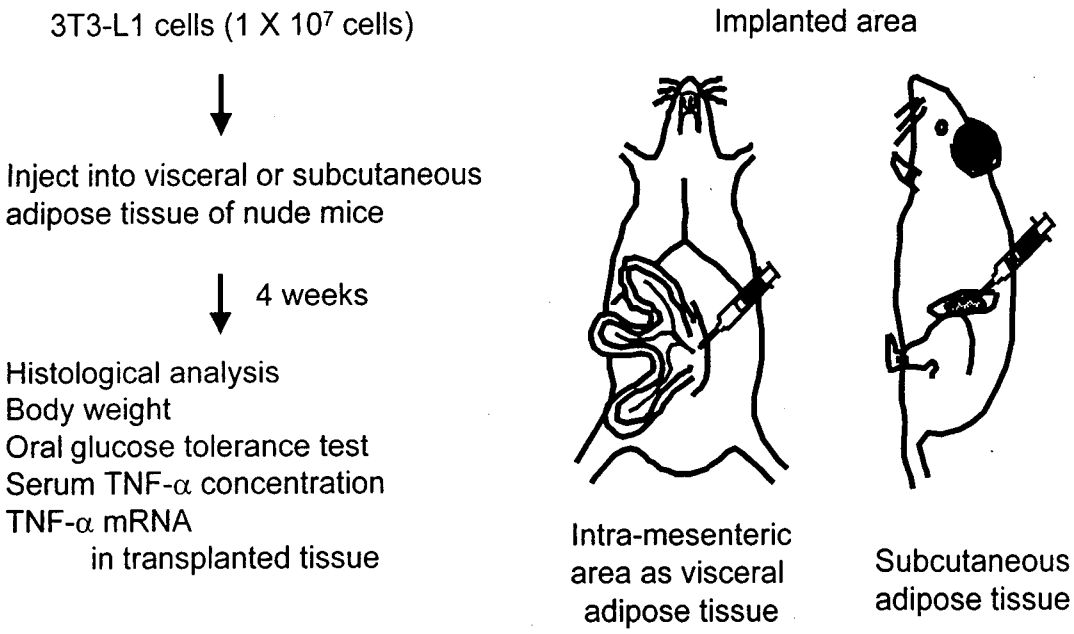

Figure 1. Implantation of adipocytes into visceral or subcutaneous adipose tissue.

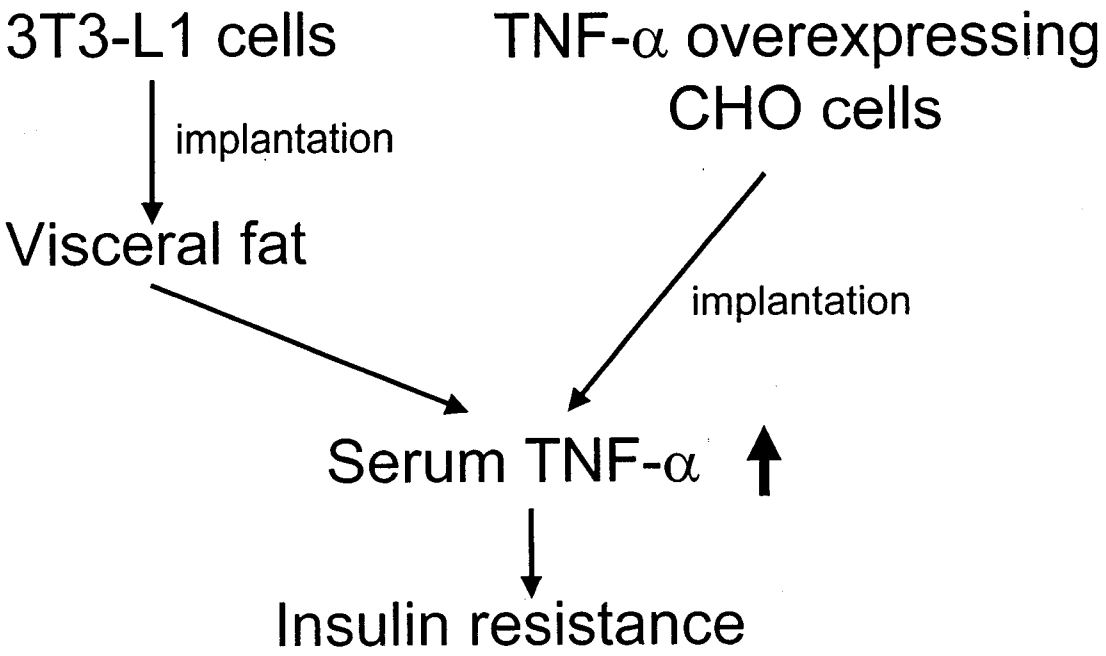

Figure 2. Functional significance of visceral fat accumulation for TNF-α-induced insulin resistance.

OGTT was analyzed at four weeks after implantation. The glucose concentrations did not show any difference between the mice implanted with 3T3-L1 cells into mesenteric area and control. During the course of glucose loading, the mice implanted with 3T3-L1 cells into mesenteric area showed remarkably increased serum insulin concentrations, compared to these of control, whereas not into subcutaneous fat area. Moreover, serum insulin concentrations of the mice, implanted with TNF-α overexpressing cells into subcutaneous fat area, were apparently higher than that of control. Furthermore, the glucose and insulin ratios at 15 minutes after loading were negatively correlated with the serum concentrations of TNF-α. These results indicate that the insulin sensitivity in mice is possibly determined by the serum TNF-α concentration caused by the secretion from the implanted cells.

The lipid profile in the serum showed the significant increase in TG concentrations of mice implanted into the mesenteric area, compared to those of mice implanted into subcutaneous area and control mice, although the concentrations of TC and HDL-C did not. Post-heparin LPL mass was negatively correlated with serum concentrations of insulin and TNF-α in the mice implanted with 3T3-L1 cells into mesenteric area. Post-heparin LPL activity was negatively correlated with insulin and TNF-α. Major sources of LPL expression were muscle and adipose tissues. Thus, the LPL expression in muscle and adipose tissues seems to be dependent on the serum insulin and TNF-α concentrations regulated by the implanted 3T3-L1 preadipocytes in mice. These results suggest that implanted preadipocytes function affected by the surrounding conditions, and the secreted TNF-α is the key regulator of the metabolic disorders such as hypertriglyceridemia accompanied with insulin resistance.

Next, the TNF-α overexpressing CHO cells were implanted into the back of nude mice subcutaneously. The serum concentrations of hTNF-α increased gradually after implantation into subcutaneous fat area. OGTT did not show any difference of the serum glucose concentrations between the mice implanted with hTNF-α overexpressing cells into subcutaneous area and control. However, the serum insulin concentrations during the course of glucose loading were apparently higher in the mice implanted with TNF-α overexpressing cells than control. Furthermore, the glucose and insulin ratios at 15 min after loading were negatively correlated with the serum TNF-α concentrations.

These results indicate that the insulin sensitivitiy in mice is possibly determined by the serum TNF-α concentration caused by the secretion from the implanted cells. Moreover, the glucose and insulin concentrations did not show any difference between the mice implanted with TNF-α overexpressing cells into subcutaneous or visceral area. These results show that TNF-α regulates the insulin sensitivity in mice even the producing cells are in subcutaneous area.

Taken together, we showed in these studies that TNF-α is a key regulator for insulin sensitivity, which accompanied with metabolic disorders. The unique method of implantation of adipose cells into subcutaneous or visceral fat area showed high TNF-α concentration and insulin resistance by the adipose cells in visceral area of nude mice (Figure 2). Furthermore, the functional significance of visceral fat accumulation for TNF-α-induced insulin resistance is partly caused by the interaction of adipocytes with surrounding conditions in mesenteric area.

On the other hand, adipose tissue expresses and releases various secretary molecules, such as leptin, plasminogen activator inhibitor-1 (PAI-1), and interleukin-6

(IL-6), in addition to TNF-α. These are potentially important to develop obesity-accompanied metabolic disorders and cardiovascular diseases. The serum levels of these cytokines are shown to be directly related to the degree of obesity of the subjects, suggesting that the circulating concentrations of these cytokines could reflect, at least in part, production by adipose tissue. A recent study showed that circulating VEGF levels were increased in the patients with coronary artery disease compared to controls asymptomatic for atherosclerosis. We therefore investigated the role of fat accumulation and its effect on serum VEGF concentrations in humans. In addition, we examined the biological activity of circulating VEGF to enhance the gene expressions of matrix metalloproteinase-3 (MMP-3) and transcriptional factor E26 transformation-specific-1 (ETS-1), which have been reported to be induced by VEGF, in endothelial cells.

Serum VEGF concentrations were positively correlated with BMI and visceral fat area. Stepwise regression analysis showed visceral fat area as the most important determinant factor. Moreover, the increased VEGF concentrations decreased as well as visceral fat area after body weight reduction. The semi-purified VEGF protein from the serum enhanced expressions of transcriptional factor Ets-1 and matrix metalloproteinase-3 in human aortic endothelial cells.

These analyses showed that the VEGF secretion from adipose tissues, particularly from intra-abdominal adipose tissues, might regulate serum concentration of VEGF (Figure 3). Increased serum VEGF concentrations associated with visceral fat accumulation could influence vascular endothelial function. Further investigations are needed to discover the molecular mechanism of increased serum VEGF concentrations accompanied with intra-abdominal fat accumulation and its pathological significance in

relation to accelerated vascular disease accompanied with obesity.

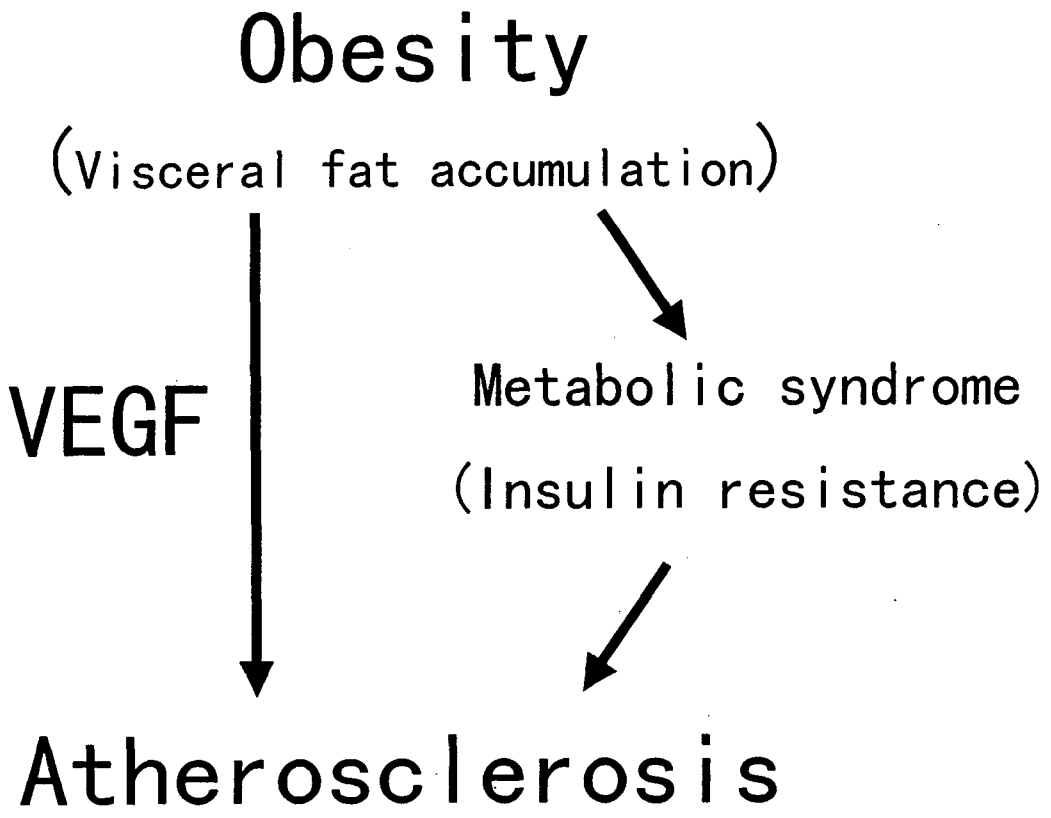

Figure 3. Increased serum VEGF concentrations associated with visceral fat accumulation could influence the progression of atherosclerosis.