The Expression of *Obese (ob)* Gene in Adipose Tissue from Wistar-Kyoto Rats (WKY) and Spontaneously Hypertensive Rats (SHR).

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One of the causes of hypertension is hyperinsulinemia. Spontaneously hypertensive rats (SHR), model rats for essential hypertension, have been reported to have hyperinsulinemia. Recently, the *obese (ob)* gene, the mutation of which results in severe hereditary obesity and diabetes in mice, has been isolated through positional cloning. The *ob* gene encodes a 166-/167-amino-acid polypeptide with a putative signal sequence, and expression of the *ob* gene occurs abundantly and specifically in the adipose tissue. We had isolated a full-length rat *ob* complementary DNA (cDNA) and demonstrated the *ob* gene expressed in adipose tissue in a region-specific manner. Since, the *ob* gene product, the OB protein (leptin) has been implicated in the regulation of body weight and fat deposition through effects on metabolism and appetite, the leptin may play an important role in hyperinsulinemia.

In this study, to elucidate the role of *ob* gene in hypertension, we analyzed the expression of the *ob* gene in the adipose tissue from SHR and normotensive control, Wistar-Kyoto rats (WKY).

Seven-week-old WKY/ Izm and SHR-B2/ Izm rats (n=3, respectively) were used in this study. They were housed in a temperature- and light-controlled room with free access to water and standard rat chow (Funabashi, SP). Food was removed from cages at 18:00 of the night preceding sacrifice. Systolic blood pressure was measured by tail cuff with a pneumatic sensor for the arterial pulse. A 0.2-ml blood was sampled from the inferior vena, and serum insulin levels were measured. Total RNA was prepared by TRIzol Reagent (GIBCO, USA) from the subcutaneous and mesenteric adipose tissue. The radioactivities of the hybridizing bands were quantitated by the image-analyzer, BAS2000 (Fuji Film, Japan). *Ob* messenger RNA (mRNA) levels were normalized by the 18S ribosomal RNA levels and were presented by arbitrary units. All data were examined by Student's *t* test for unpaired observation. Data are presented as means $\pm$ SEM.

In seven-week-old SHR, systolic blood pressure tended to higher than WKY (138.7 ± 4.3 vs. 123.0 ± 1.5 mmHg, respectively). As has been previously reported, SHR weighted less than WKY (126.3 ± 1.1 vs. 138.4 ± 7.0 g; $p < 0.05$). Fasted serum insulin in SHR was significantly higher than that of WKY (1.65 ± 0.40 vs. 0.57 ± 0.03 ng/ml; $p < 0.05$).

Northern blot analysis using the cloned rat *ob* cDNA as a probe identified a single mRNA species of 4.5 kb in size in the adipose tissue with regional specificities. The *ob* mRNA level in mesenteric fat from SHR was approximately two times higher than that from WKY (1.76 ± 0.43 vs. 0.97 ± 0.25). On the other hand, the levels of *ob* mRNA in subcutaneous fat was near the same between WKY and SHR (2.26 ± 0.29 vs. 2.39 ± 0.25).

The present results demonstrated that the *ob* gene expression was augmented in the mesenteric adipose tissue from SHR as compared with that from WKY, while the *ob* mRNA level in the subcutaneous adipose tissue from SHR were almost the same as that from WKY, and the serum insulin level in SHR was significantly higher than in WKY. Further studies are required for the elucidation of the relationship between the augmented *ob* gene expression and the high serum insulin level in SHR.