Tumor Necrosis Factor-α Levels in Adipose Tissue of Lean and Obese Cats\textsuperscript{1,2}

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EXPANDED ABSTRACT

KEY WORDS: • TNFα • feline • adipose tissue • FIHL • ELISA

Obesity has traditionally been thought of as a disease state based on a chronic excess of energy intake compared with energy expenditure. Until recently, the adipocyte was thought to be a passive cell for the development of obesity state. Current advances in obesity research have led to the discovery that adipocytes have a regulatory role. Although adipocyte enlargement and hypertrophy comprise obesity onset, it appears that this cell also has an endocrine role and participates in metabolic pathways that result in an obese state. Previously it was proposed that the adipocyte may serve as an “adipostat” by releasing a signal to help control food intake and regulate a body fat “set point” in each individual. However, because an adipocyte-derived signal was not discovered, interest in the regulatory role of the adipocyte lessened (Hotamisligil et al. 1993, Hotamisligil and Speigelman 1994). Recently, the discovery of various molecules produced and secreted by the adipocyte has renewed interest in this area. These compounds include adipin (complement factor D), angiotensinogen and tumor necrosis factor-α (TNFα).\textsuperscript{4}

TNFα, or cachectin, is a cytokine normally associated with the acute phase response. It is produced by macrophages in response to inducers such as endotoxin, inflammation and cancer. The discovery that TNFα is produced by adipocytes stemmed from investigations into the regulation of complement proteins (adipsin) in adipocytes. Perhaps most remarkable is the observation that obese human beings and rodents have significantly increased TNFα expression (Flier 1995, Hotamisligil et al. 1993, Hotamisligil and Speigelman 1994). Other research has suggested that TNFα may be involved in the anorexia associated with anorexia nervosa (Vaisman and Hahn 1991). Recent reports have indicated a role for TNFα in the insulin resistance of noninsulin-dependent diabetes mellitus (Hotamisligil et al. 1993, Hotamisligil and Speigelman 1994). TNFα has also been shown to cause a suppression of most lipogenic enzymes, including adipose tissue lipoprotein lipase (LPL). Elevated levels of TNFα are commonly seen in genetic mouse models of insulin resistance (Hotamisligil et al. 1993), whereas increased levels of other cytokines were not observed in insulin-resistant obese mouse models. The insulin resistance effect by TNFα is thought to be mediated in part by an ability to inhibit intracellular signaling of tyrosine kinase from the insulin receptor (Hotamisligil et al. 1995). Direct evidence exists indicating a role in human beings for TNFα in obesity-induced insulin resistance. Both TNFα mRNA and corresponding protein levels increase \textgreater2.5-fold in obese adults compared with age matched controls (Hotamisligil et al. 1995). Weight loss in obese individuals who regain insulin sensitivity leads to significant reductions in adipose tissue TNFα levels; however, circulating levels of TNFα do not change (Hotamisligil et al. 1995).

An inverse relationship exists between TNFα expression and LPL activity in obese human beings. TNFα appears to inhibit LPL activity in adipose tissue, and this trend is reversed when these same subjects lose weight. The relationship between TNFα and LPL is also thought to mediate some of the insulin resistance seen in obese patients (Kern et al. 1995). Based on observations such as these, TNFα has been proposed to be an adipocyte regulator to aid in obesity regulation in some human subjects. Metabolic manipulation of TNFα levels and LPL activity may offer ways to aid in obesity and obesity-related disease therapy.

The incidence of obesity in pet animals is astounding. Some figures estimate that 50% of cats living indoors are obese and 30% of cats with access to outdoor activities are obese (Sloth 1992). Obese animals tend to have shortened life spans and a predisposition to other medical problems such as circulatory, locomotive, dermatologic, reproductive and neoplastic diseases. In cats, the disease idiopathic feline hepatic lipidosis (IFHL) is associated with obesity, anorexia and disturbances in lipid metabolism. Because of this correlation, we hypothesized that TNFα may be involved in the etiology of IFHL. An ability to modulate obesity and obesity-related diseases by

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\textsuperscript{4} Abbreviations used: BMI, body mass index; IFHL, ideopathic feline hepatic lipidosis; LPL, lipoprotein lipase; TNF, tumor necrosis factor.
altering adipocyte regulators may help facilitate therapy and prevention. Based on work done both in human beings and rodents, we hypothesized that adipose tissue TNFα levels in obese cats would be elevated compared with levels in lean cats.

**Materials and methods.** Twelve adult neutered male and female cats obtained from the University of Georgia Animal Resources were divided into two groups on the basis of body mass index (BMI). Animals were housed and cared for in accordance with the NIH Guide for the Care and Use of Laboratory Animals. BMI was determined by dividing the body weight (kg) of the cat by the product of height × length (m²). The obese cat group had a BMI >42, whereas the lean group had a BMI <42. Abdominal fat biopsies (subcutaneous adipose tissue) were obtained from each cat using local anesthesia. Adipose tissue samples were homogenized (PowerGen 700D Model, Fischer Scientific, Atlanta, GA) in PBS, centrifuged and supernatants were collected. Feline adipose tissue samples were homogenized (PowerGen 700D Model, Fischer Scientific, Atlanta, GA) in PBS, centrifuged and supernatants were collected. Feline homogenizers 700D Model, Fischer Scientific, Atlanta, GA) in PBS, centrifuged and supernatants were collected. Feline adipose tissue samples were homogenized (PowerGen 700D Model, Fischer Scientific, Atlanta, GA) in PBS, centrifuged and supernatants were collected. Feline adipose tissue samples were homogenized (PowerGen 700D Model, Fischer Scientific, Atlanta, GA) in PBS, centrifuged and supernatants were collected. Feline adipose tissue samples were homogenized (PowerGen 700D Model, Fischer Scientific, Atlanta, GA) in PBS, centrifuged and supernatants were collected.

**Results and discussion.** Adipose tissue samples were weight matched, and lean and obese cat groups were expressed on the basis of body weight and body mass index. Lean cats had undetectable levels of TNFα, whereas obese cats had 1.79 pg/kg body weight (Table 1). On the basis of body mass index, obese cats had 0.19 pg/(kg·m²) TNFα, whereas levels in lean cats were below the level of assay sensitivity (Table 1). These results were expected on the basis of work performed on both human beings and rodents that showed higher level of TNFα protein and TNF mRNA in obese subjects compared with lean subjects (Hotamisligil et al. 1993, Kern et al. 1995). As in those studies, we did not find any TNFα in cat serum.

We also investigated TNFα levels in adipose tissue of two cats with clinically confirmed IFHL. Both cats were anorexic (~2 wk) upon diagnosis and initial fat biopsy. A gastrostomy feeding tube was then placed in each cat and nutritional therapy was started. By d 7, adipose tissue TNFα levels decreased to levels comparable to those of clinically normal obese cats (Fig. 1). This effect may have been due to a catabolic state induced by the anorexia. However, it is also possible that the decrease may have been due to a regulatory effect exerted by adipose tissue TNFα. If this were true, then TNFα may have a useful role in the diagnosis, treatment, and/or prevention of IFHL and perhaps other obesity-related diseases. Also, because obesity and increased levels of adipose TNFα can be correlated with insulin resistance, it will be interesting in future studies to measure insulin sensitivity in obese cats.

### TABLE 1

<table>
<thead>
<tr>
<th>Cats</th>
<th>TNFα</th>
<th>TNFα</th>
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<tbody>
<tr>
<td></td>
<td>pg/kg body weight</td>
<td>pg/(kg · m²)</td>
</tr>
<tr>
<td>Obese</td>
<td>1.79 ± 0.30</td>
<td>0.19 ± 0.04</td>
</tr>
<tr>
<td>Lean</td>
<td>ND¹</td>
<td>ND</td>
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</tbody>
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¹ ND, below sensitivity level of assay.

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**REFERENCES**


