

Leptin, adiponectin levels, and thyroid hormones in normal and obese dogs

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Abstract : The present study compared leptin, adiponectin, and thyroid hormone concentrations in normal and obese dogs, and evaluated the association between leptin and adiponectin concentrations and thyroid function. The serum leptin, adiponectin, thyroid-stimulating hormone (TSH), total thyroxine (tT4), free thyroxine (fT4), triiodothyronine (T3), and cortisol concentrations were measured in 18 normal dogs (body condition score [BCS]: 4-5/9) and 16 obese dogs (BCS: 8-9/9). Leptin and T3 concentrations were higher in the obese group than the normal weight group ($p < 0.01$ and $p < 0.05$, respectively). In both groups, the T3 and leptin concentrations were correlated ($r = 0.370$, $p < 0.05$), as were the TSH and fT4 and adiponectin concentrations ($r = -0.373$, $p < 0.05$ and $r = 0.369$, $p < 0.05$, respectively). In the normal weight group, the TSH and fT4 concentrations were correlated with the adiponectin concentrations ($r = -0.528$, $p < 0.05$ and $r = 0.482$, $p < 0.05$, respectively). The results of the present study suggest that leptin and T3 concentrations are significantly higher in obese dogs than normal weight dogs, and the serum T3 and leptin concentrations are positively correlated.

Keywords : adiponectin, leptin, obese dog, thyroid hormones

Introduction

Obesity is a condition characterized by excessive adipose tissue accumulation in the body and is a common nutritional problem in small animal medicine [16]. In addition to its role as a repository of excessive energy, adipose tissue has become recognized as a metabolically active organ that secretes various adipokines such as leptin and adiponectin. Evidence suggests that adipokines may contribute to numerous regulatory processes, including inflammation and immune function [25], hemostasis and vascular function [9], hematopoiesis [3], and cell proliferation and angiogenesis [27].

Plasma leptin concentration is positively correlated with body fat mass in dogs, consistent with findings in humans and rodents [13]. In contrast to leptin, circulating adiponectin concentration is negatively correlated with fat mass, and weight loss leads to an increased adiponectin concentration in humans, primates, and rodents [12]. Adiponectin is best known for its ability to enhance insulin sensitivity, counteract inflammation, and inhibit atherosclerosis development [11].

Hypothyroidism is a commonly cited cause of obesity. The prevalence of hypothyroidism in dogs is an estimated 0.2%, and less than half of these dogs are obese [8, 21]. The prev-

alence of canine obesity is far higher (25~40%) [18], indicating that obesity should be considered a nutritional problem rather than a secondary condition in many cases. In human studies, obesity has been shown to influence thyroid homeostasis [23, 24], and disturbances in thyroid function can similarly occur in obese dogs. Weight loss may lead to decreased total triiodothyronine (T3) and thyroid-stimulating hormone (TSH) concentrations [2]. Therefore, adipokines, such as leptin and adiponectin, may also affect thyroid hormones function. Several studies reported a relationship between adipokines and thyroid hormones in humans and rodents [1, 22]. In dogs, one study reported an association between canine hypothyroidism and increased serum leptin concentration [17].

The present study compares leptin, adiponectin, and thyroid hormone concentrations in normal and obese dogs.

Materials and Methods

Subjects

In total, 34 dogs were divided into two groups: 18 were assigned to the normal weight group (body condition score [BCS]: 4-5/9), and 16 were assigned to the obese group (BCS: 8-9/9). The BCS and gender are summarized in Table 1.

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Table 1. Distributions for body condition score (BCS) and gender in normal and obese dogs

	Normal	Obese
BCS		
4	12	–
5	6	–
7	–	–
8	–	7
9	–	9
Gender		
Intact male	1	1
Castrated male	6	4
Intact female	10	8
Spayed female	1	3

The dogs were aged from 2 to 11 years, with no significant difference between the two groups. The normal group included Maltese (n = 3), Poodle (n = 2), Schnauzer (n = 2), Shih Tzu (n = 2), and mixed breeds (n = 9). The obese group included a Beagle (n = 1), Chihuahua (n = 1), Maltese (n = 2), Miniature Pincher (n = 1), Pomeranian (n = 2), Poodle (n = 2), Shih Tzu (n = 4), Welsh corgi (n = 1), and Yorkshire terrier (n = 2). Each dog underwent a physical examination, as well as serum biochemistry and electrolyte analyses, and the BCS was assessed on a 9-point scale.

Subjects and sample collection

The age, gender, breed, and clinical history of 34 client-owned dogs were recorded. The dogs were examined at the Veterinary Medical Teaching Hospital for routine physical examinations and were all deemed healthy. The dogs were fasted for 12 hours prior to blood collection. Blood was collected from the cephalic vein, allowed to clot at room temperature for 30 minutes, and then centrifuged. Serum aliquots were stored at -80°C in plastic tubes until measurement of the leptin, adiponectin, TSH, total thyroxine (tT4), free thyroxine (fT4), T3, and cortisol concentrations.

Sandwich enzyme-linked immunosorbent assay and hormone assay

Serum leptin and adiponectin concentrations were measured according to the manufacturer's protocol using a canine leptin commercial sandwich enzyme-linked immunosorbent assay (ELISA) kit (Millipore, USA) and canine adiponectin commercial ELISA kit (LINCO Research, USA). Serum TSH, tT4, fT4, T3, and cortisol concentrations were quantified using an Immulite 1000 immunoassay analyzer (Siemens Healthcare, USA).

Statistical analysis

The normal and obese groups were compared according to gender using the Chi-squared test. All data, except the breed and gender, are expressed as mean \pm standard error of the

Table 2. Serum chemistries and electrolytes in normal (n = 18) and obese (n = 16) dogs

	Normal	Obese	P value
ALP (U/L)	81.06 \pm 12.20	117.44 \pm 15.55*	0.027
AST (U/L)	27.78 \pm 2.53	20.21 \pm 3.24**	0.009
BUN (mg/dl)	20.74 \pm 1.25	16.49 \pm 2.08*	0.027
TP (g/dl)	5.94 \pm 0.23	6.21 \pm 0.14*	0.043
Sodium (mEq/L)	142.92 \pm 1.01	146.39 \pm 0.58*	0.014

* $p < 0.05$; ** $p < 0.01$, vs. normal (mean \pm SE). ALP: alkaline phosphatase, AST: aspartate aminotransferase, BUN: blood urea nitrogen, TP: total protein.

Table 3. Adipokines, TSH, thyroid hormones and cortisol in normal (n = 18) and obese (n = 16) dogs

Hormones	Normal	Obese	P value
Leptin (ng/mL)	3.55 \pm 0.65	10.85 \pm 2.14**	0.001
Adiponectin (ng/mL)	8.42 \pm 1.04	7.53 \pm 1.04	0.388
TSH (ng/mL)	0.14 \pm 0.40	0.14 \pm 0.41	0.986
tT4 ($\mu\text{g/dL}$)	1.47 \pm 0.15	1.33 \pm 0.17	0.569
fT4 (ng/dL)	1.65 \pm 0.09	1.63 \pm 0.15	0.769
T3 (ng/dL)	66.55 \pm 4.10	80.81 \pm 4.64*	0.038
Cortisol ($\mu\text{g/dL}$)	4.84 \pm 0.68	4.96 \pm 0.85	0.945

* $p < 0.05$; ** $p < 0.01$, vs. normal (mean \pm SE). TSH: thyroid-stimulating hormone, tT4: total thyroxine, fT4: free thyroxine, T3: triiodothyronine.

mean (SEM). Age, BCS, and the leptin, adiponectin, TSH, tT4, fT4, T3, and cortisol concentrations were compared between the groups using the Mann-Whitney test. Associations between the variables were analyzed using the Spearman correlation test. All of the analyses were performed using PASW Statistics 18 software (SPSS, USA).

Results

Gender distribution was similar between the two groups. Serum alkaline phosphatase (ALP), total protein, and sodium were higher in obese dogs than in normal dogs, and aspartate transaminase (AST) and blood urea nitrogen (BUN) were lower in obese dogs than in normal dogs (Table 2). The leptin, adiponectin, TSH, tT4, fT4, T3, and cortisol concentration are shown in Table 3. Leptin concentrations were not measured in four dogs (3 spayed and 1 intact female) in the obese group due to an assay error. Leptin and T3 were significantly higher in the obese group than in the normal group ($p < 0.01$ and $p < 0.05$, respectively). Serum TSH, tT4, and fT4 concentrations showed no significant difference between the normal and obese groups.

T3 and leptin concentrations were correlated ($r = 0.370$, $p < 0.05$; Fig. 1). TSH and fT4 concentrations were correlated with adiponectin concentration ($r = -0.373$, $p < 0.05$ and $r = 0.369$, $p < 0.05$, respectively; Fig. 2). In the normal

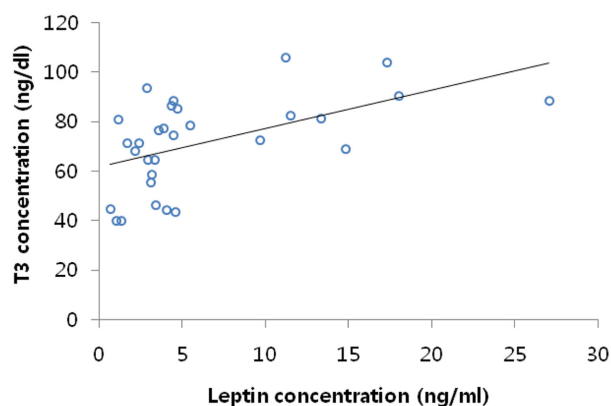


Fig. 1. Relationship of leptin with T3 in all dogs of both groups. Serum T3 level positively correlated with leptin ($n = 34$, $r = 0.370$, $p < 0.05$).

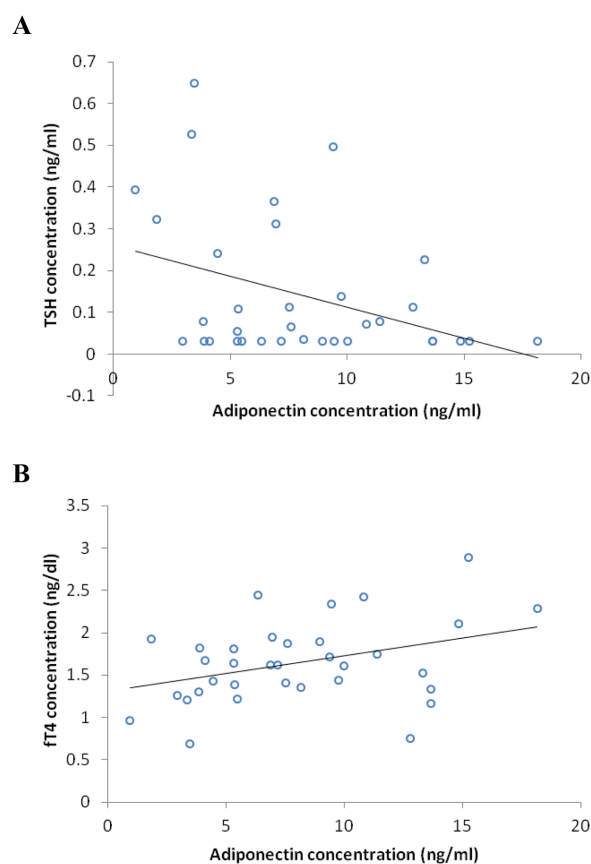


Fig. 2. Relationship of adiponectin with thyroid hormones in all dogs of both groups. (A) Correlation of serum adiponectin levels with TSH ($n = 34$, $r = -0.373$, $p < 0.05$). (B) Correlation of serum adiponectin levels with fT4 ($n = 34$, $r = 0.369$, $p < 0.05$).

group, TSH and fT4 concentrations were correlated with adiponectin concentrations ($r = -0.528$, $p < 0.05$ and $r = 0.482$, $p < 0.05$, respectively; Fig. 3). No correlations were found between the remaining variables.

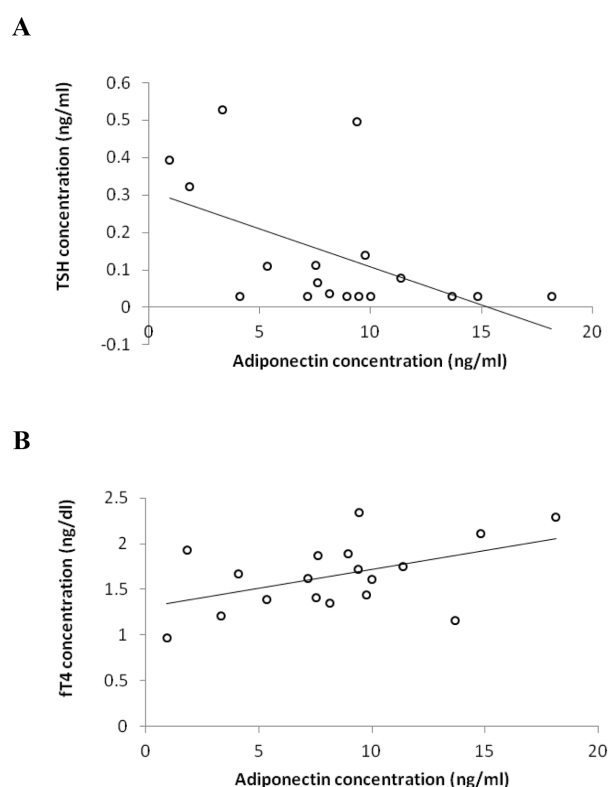


Fig. 3. Relationship of adiponectin with thyroid hormones in the normal body weight group. (A) Correlation of serum adiponectin levels with TSH ($n = 18$, $r = -0.528$, $p < 0.05$). (B) Correlation of serum adiponectin levels with fT4 ($n = 18$, $r = 0.482$, $p < 0.05$).

Discussion

Many dogs are diagnosed with and treated for hypothyroidism in veterinary practice because they are overweight. However, obesity is a common nutritional condition in dogs, and many obese dogs do not have concurrent hypothyroidism [15]. The present study showed that serum leptin and total T3 concentrations were higher in obese dogs than in normal dogs. These results are consistent with previous human and canine studies. In one study, the leptin concentration was higher in obese people than in lean people [22]. Similarly, several canine studies reported higher leptin concentrations in obese dogs than in lean dogs [13]. Obesity or weight gain is also associated with elevated T3 concentrations in dogs [2] and humans [23]. Unconfirmed observations suggest that the increased T3 concentration in obese individuals is caused by an increased conversion of T4 to T3 and decreased T3 receptors [8]. Adiponectin, TSH, tT4, and fT4 concentrations may also be associated with obesity in humans [8, 25], rhesus monkeys [12], and dogs [2, 4].

An association between adipokines and thyroid hormones has been reported in humans and rodents [1, 22], but not in dogs. The present study demonstrated that T3 and leptin concentrations are positively correlated. A rodent study revealed

that the type 2-deiodinase mRNA expression and activity in brown adipose tissue markedly increased following systemic leptin administration, likely causing an increased T3 and decreased T4 plasma concentrations [1]. Another rodent study found that systemically administered leptin stimulated fT3 and tT4 release through a direct mechanism involving Ob-Rb and inhibited TSH release [20]. In humans, leptin and T3 concentrations were correlated in obese humans receiving a long-term, low-calorie diet, and both hormones reportedly decreased. Potentially, the T3 concentration may decrease in response to the negative energy balance caused by a low-calorie diet [19]. Therefore, the increased leptin concentration necessary for energy expenditure may be followed by changes in the T3 concentration in obese individuals.

In the present study, TSH and adiponectin concentrations were negatively correlated, and fT4 and adiponectin concentrations were positively correlated. Two studies in humans reported the reverse in the association between TSH and adiponectin concentrations. One study reported that TSH and adiponectin concentrations were inversely correlated in severely obese women [14], which is consistent with the present findings. These results suggest that TSH is positively associated with obesity severity and may be an early marker of altered energy balance in obese individuals [14]. A positive correlation was found between fT4 and adiponectin concentrations, consistent with a human study that showed a higher serum fT4 concentration in healthy subjects with high adiponectin concentrations [5]. Other studies suggest that adiponectin may regulate thyroid hormone production via gC1q receptors within thyroid mitochondria [26]. However, another study suggested that the association between adiponectin and thyroid hormones could be indirect and occur through insulin sensitivity or cold exposure [21]. Cortisol concentrations were also evaluated to exclude any cortisol effects on the thyroid hormones. In the present study, there was no correlation between cortisol and thyroid hormone concentrations.

The ALP concentration was higher in obese dogs, consistent with previous studies [4, 7]. Higher total protein and sodium concentrations, and lower BUN and AST concentrations were also found in the obese dogs, although the clinical significance of these differences is unknown. Ten castrated male and 4 spayed female dogs were included in the present study. Gonadectomy does not generally affect thyroid or adrenocortical function, although slight differences between gonadectomized and intact dogs have been reported [6, 10]. The mechanism underlying the adipokine and thyroid hormone associations in canines requires further study.

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