

Influence of obesity on plasma lipid and lipoprotein concentrations in dogs

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Objective—To determine effects of obesity and diet in dogs on plasma lipid and lipoprotein concentrations by assaying plasma leptin and ghrelin concentrations and determining total plasma cholesterol and triglyceride concentrations as well as the concentrations of cholesterol and triglycerides in various lipoprotein classes (ie, very-low-density, low-density, and high-density lipoproteins).

Animals—24 Beagles; 12 lean (mean \pm SEM) body weight, 12.7 ± 0.7 kg) and 12 chronically obese (21.9 ± 0.8 kg) dogs of both sexes, between 1 and 9 years old.

Procedures—Total plasma cholesterol and triglyceride concentrations; lipoprotein cholesterol and triglyceride concentrations; and plasma ghrelin, leptin, free fatty acids, insulin, and glucose concentrations were measured and compared between lean and obese dogs, both of which were fed a complete and balanced maintenance diet. Chronically obese dogs were subsequently fed a high-protein low-energy diet to evaluate effects of diet composition on plasma lipid and lipoprotein measurements.

Results—Chronic obesity resulted in a significant decrease in plasma ghrelin concentration and a significant increase in plasma leptin, cholesterol, and triglyceride concentrations in dogs. High total plasma cholesterol and triglyceride concentrations resulted from increased cholesterol and triglyceride concentrations in all lipoprotein fractions. In obese dogs, modification of diet composition resulted in beneficial effects on plasma lipid and leptin concentrations, even before weight loss was observed.

Conclusions and Clinical Relevance—Correlations exist between obesity and plasma measurements (ie, lipoproteins, leptin, insulin, and ghrelin) commonly associated with obesity. Modification of diet composition to control energy intake improves plasma lipid and leptin concentrations in obese dogs. (*Am J Vet Res* 2005;66:81–86)

Obesity and excess body weight (BW) are the most common nutritional disorders encountered in small animal medicine. They are estimated to affect approximately 25% to 44% of dogs receiving veterinary care in Western countries.^{1,2} Obesity develops when energy intake consistently exceeds daily energy expenditure. Numerous environmental and social factors

contribute to the formation of obesity. These include lack of exercise, overfeeding, or unbalanced diet. Ad libitum feeding of a high-fat diet, for example, is a well-known factor of obesity development.³ Genetic factors such as breed or physiologic factors such as neutering have been associated with an increase in the risk of obesity. Drugs (eg, corticosteroids and progestones) or endocrine abnormalities have also been implicated as causes of obesity.

Cholesterol and triglycerides are transported through plasma in special particles called lipoproteins. In canine plasma, Mahley and Weisgraber⁴ identified lipoproteins with the physical and chemical characteristics of very-low-density lipoprotein (VLDL), low-density lipoprotein (LDL), and high-density lipoprotein (HDL). Hyperlipidemia, a term used to describe an increase in plasma concentrations of cholesterol, triglycerides, or both, is caused by defects in the metabolism of ≥ 1 of the lipoprotein classes that may be either genetic in origin or, more commonly in dogs, secondary to diseases. In dogs as in humans, obesity or high-fat diets are associated with insulin resistance. In humans, insulin resistance is associated with hyperlipidemia. Dogs are currently used to study insulin resistance and associated dyslipidemia.⁵

Leptin is a protein synthesized and secreted primarily by adipose tissue. Circulating leptin concentrations are high in obese dogs.⁶ Moreover, it is suggested that plasma leptin is a quantitative marker of adiposity in dogs.^{6,7} Ghrelin, a gastric peptide, was identified in dogs by Tomatesso et al.⁸ Canine ghrelin has a high homology with rodent and human ghrelin. In addition to its growth-hormone releasing properties, exogenous ghrelin injections stimulate food intake and adiposity in humans and rodents.^{9–13} In obese humans, plasma ghrelin concentrations are low.^{11–13}

Evaluation of the effect of obesity on the measurement of blood lipids and lipoproteins is important because hyperlipidemia can result in interference with laboratory measurements. Hypertriglyceridemia interferes with direct bilirubin determination in serum, resulting in moderate increases in concentration. Hypertriglyceridemia often results in decreases in serum cholesterol, chloride, amylase, and lipase concentration measurements and directly interferes with plasma protein and hemoglobin assays. Hypercholesterolemia may lower measured triglyceride concentrations.¹⁴

The purposes of the study reported here were to assay plasma leptin and ghrelin concentrations and determine total plasma cholesterol and triglyceride concentrations, as well as the concentrations of cholesterol and triglyceride in various lipoprotein classes (ie, VLDL, LDL, and HDL) in lean and obese dogs under

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controlled experimental conditions. In humans, the health benefits of diets low in fat and rich in complex carbohydrates on blood lipid concentrations are well recognized. In the present study, effects of a low-energy high-protein low-fat diet in obese dogs were also studied.

Material and Methods

Animals and diet—Twenty-four Beagles (12 control and 12 obese dogs) between 1 and 9 years old, housed at the Animal Nutrition Unit, University of Liège, Belgium, were used for this study. Groups consisted of 3 sexually intact females, 3 spayed females, and 6 neutered males. All dogs were assessed as healthy (except for obesity) on the basis of findings of physical examination, CBC determination, and serum biochemical analysis. Dogs were maintained in pairs in their usual kennels during the study. Each pen was a 4 × 3-m size and contained a doghouse. Dogs were separated and fed a complete and balanced maintenance commercial extruded (dry-type) diet^a (Appendix) individually once a day in the morning for 90 minutes. Drinking water was provided ad libitum. Activity levels of dogs were not recorded. Body weights were recorded once a week before feeding. Body condition score was assessed in each group according to a validated 9-point body condition scoring system.¹⁵

For lean control-group dogs, mean (\pm SEM) BW was 12.7 ± 0.7 kg, body condition score of all dogs was 5 on the 9-point scale, and mean age was 4.4 ± 0.9 years old. For obese-group dogs, mean BW was 21.9 ± 0.8 kg (48% overweight), body condition scores were 7 or 8, and mean age was 4.7 ± 0.6 years old. Obesity had been induced in obese-group dogs by feeding a maintenance diet^a in large quantities during a period of 10 to 15 months. Food left uneaten after 90 minutes was collected and weighed. Dogs were grossly obese with a stable BW for at least 1 year at the time of the evaluation. Individual intake was recorded daily for the last month to determine a baseline value.

The 12 obese-group dogs were then fed a high-protein low-energy commercial extruded (dry-type) diet^b (Appendix) for 1 month. To adapt obese-group dogs to their new diet, they were offered equal amounts of the 2 diets mixed together for 1 week and then exclusively the new diet for the remaining weeks. The amount of the low-energy diet fed to each obese-group dog was calculated. The purpose was to offer each dog the same energy content as in the baseline diet. If food was left uneaten after 90 minutes, it was collected and weighed. The Animal Use and Care Advisory Committee of the University of Liège, Belgium, approved the protocol.

Sample collection—Blood samples from all dogs were collected from the cephalic vein. All dogs were fed their diet for at least 1 month before blood sample collection. Blood samples from all dogs were collected between 9 and 10 AM after 24 hours of withholding food. Blood samples were collected into tubes containing potassium EDTA for analysis of cholesterol and triglyceride concentrations in plasma and lipoproteins and for analysis of plasma concentrations of ghrelin, leptin, and nonesterified fatty acids. Blood samples for the measurement of glucose and insulin concentrations were collected into fluoride heparin tubes. Plasma was separated by low-speed centrifugation at 4°C. Plasma samples for the measurement of lipoprotein cholesterol and triglyceride concentrations were stored at 4°C and analyzed within 72 hours of collection. Plasma samples for the measurement of the other blood variables were stored in plastic tubes at -80°C until assay.

Plasma lipid and lipoprotein concentrations—Separation and quantification of plasma lipoproteins were

performed on fresh plasma by use of a combined ultracentrifugation and precipitation method, previously validated for use with canine plasma.¹⁶ Briefly, normal saline (0.9% NaCl) solution was layered over plasma and then ultracentrifuged at $164,000 \times g$, 4°C, for 18 hours. Lipoproteins with a density of < 1.006 g/mL (ie, VLDL) were recovered from the top fraction by tube slicing. Infranatant containing lipoproteins of < 1.006 g/mL was made up to a total volume of 5 mL. Apoprotein B containing lipoproteins (ie, LDL) was then precipitated by incubation and centrifugation of the infranatant with 92mM heparin-manganese chloride solution. Supernatant of this centrifugation contained HDL lipoprotein. The LDL precipitate was resolubilized with 0.5M sodium citrate and 0.15M sodium chloride and dialyzed against sodium azide. Lipoprotein fractions (ie, VLDL, LDL, and HDL) were then made up to a total volume of 3 mL with normal saline solution. Lipoprotein fractions and total plasma samples were then tested by horizontal electrophoresis on agarose lipogel^c to assess the efficiency of the separation of lipoprotein fractions. On the basis of the known characteristics of canine lipoproteins, it was assumed that chylomicrons would remain at the origin and VLDL, LDL, HDL₁, and HDL₂ would have pre- β -, β -, α_2 -, and α_1 -lipoprotein mobilities.¹⁷ Total plasma samples and lipoprotein fractions were then frozen at -80°C in plastic tubes until assayed. Cholesterol and triglyceride concentrations were measured by use of an automatic analyzer^d with a commercial kit.^{e,f} Cholesterol concentrations were measured in each fraction, whereas triglyceride concentrations were measured in total plasma samples and in the VLDL fraction. Triglyceride concentrations in LDL and HDL (LDL-HDL) fractions together were calculated by subtraction. Coefficients of variation (within assay) were calculated from 10 plasma samples of blood collected from 1 dog on a single day and then treated separately. Coefficients of variation for measurements of cholesterol concentrations were 0.59%, 4.17%, 7.2%, and 14.38% in total plasma and in VLDL, HDL, and LDL fractions, respectively. Coefficients of variation for measurements of triglyceride concentrations were 0.88% and 9.13% in total plasma and the VLDL fraction, respectively.

Plasma ghrelin, leptin, nonesterified fatty acids, insulin, and glucose concentrations—Plasma ghrelin and insulin concentrations were measured by use of commercial human radioimmunoassay kits.^{g,h} These kits may also be used with canine plasma. Concentrations of nonesterified fatty acids and glucose were determined by use of commercial kits.^{i,j} Leptin was assayed by use of a canine-specific ELISA method, previously validated for use in dogs.¹⁸ Coefficients of variation (calculated from 10 plasma samples of blood collected from 1 dog on a single day and then treated separately) for plasma total ghrelin, leptin, insulin, and glucose concentrations were 5.3%, 3.6%, 5.9%, and 1.4%, respectively.

Statistical analyses—Values are expressed as mean \pm SEM. Data were normally distributed and were subjected to an ANOVA, with group as a fixed effect.^{19,k} Groups studied were lean control-group dogs, obese-group dogs fed the complete and balanced maintenance diet, and the same obese-group dogs fed the low-calorie diet. Pearson correlation analysis was performed on data. Values of $P < 0.05$ were considered significant.

Results

Energy consumption, BW, and sex—Mean energy consumption by obese-group dogs fed the maintenance diet was significantly greater than for lean control-group dogs (Table 1). Once the obese-group dogs were fed the low-energy diet, mean energy consumption significantly decreased by 45%.

The BW of obese-group dogs was significantly

Table 1—Mean (\pm SEM) body weight (BW) and energy intake* in lean control-group and obese-group dogs fed 2 diets.

Dog groups (n)	Diet	Measurements		
		BW (kg)	Energy intake (kJ/kg of BW ^{0.75})	Energy intake (kJ/kg of TBW ^{0.75})
Lean control (12)	Maintenance	12.7 \pm 0.7	490 \pm 0	490 \pm 0
Obese (12)	Maintenance	21.9 \pm 0.8†	649 \pm 29†	840 \pm 18†
Obese (12)	Low energy	21.5 \pm 0.8†	372 \pm 17‡	463 \pm 12‡

*Measured the week of blood sample collection. †Significantly ($P < 0.05$) different from lean control-group dogs. ‡ Significantly ($P < 0.05$) different from obese-group dogs fed a complete and balanced diet. TBW = Target body weight.

Table 2—Mean (\pm SEM) plasma glucose, insulin, leptin, ghrelin, and nonesterified fatty acid concentrations in lean control-group and obese-group dogs fed 2 diets.

Dog groups (n)	Diet	Measurements				
		Glucose (mg/dL)	Insulin (pmol/L)	Leptin (ng/mL)	Ghrelin (pg/mL)	NEFA (mmol/L)
Lean control (12)	Maintenance	87.37 \pm 2.16	56.2 \pm 9.0	2.29 \pm 0.43	4,084 \pm 460	0.57 \pm 0.07
Obese (12)	Maintenance	87.37 \pm 3.24	107.2 \pm 12.7*	13.18 \pm 1.93*	2,336 \pm 150*	0.83 \pm 0.11
Obese (12)	Low energy	91.52 \pm 1.26	100.0 \pm 11.6*	9.88 \pm 1.58*†	2,950 \pm 483*	1.05 \pm 0.14*†

*Significantly ($P < 0.05$) different from lean control-group dogs. †Significantly ($P < 0.05$) different from obese-group dogs fed a maintenance diet. NEFA = Nonesterified fatty acids.

Table 3—Mean (\pm SEM) cholesterol and triglyceride concentrations in total plasma and in plasma lipoprotein fractions in lean control-group and obese-group dogs fed 2 diets.

Dog groups (n)	Diet	Measurements						
		Cholesterol (mmol/L)	VLDL-C (mmol/L)	HDL-C (mmol/L)	LDL-C (mmol/L)	Triglyceride (mmol/L)	VLDL-T (mmol/L)	LDLHDL-T (mmol/L)
Lean control(12)	Maintenance	4.47 \pm 0.28	0.10 \pm 0.03	3.88 \pm 0.23	0.49 \pm 0.05	0.60 \pm 0.08	0.32 \pm 0.07	0.28 \pm 0.03
Obese (12)	Maintenance	6.28 \pm 0.39*	0.23 \pm 0.05*	5.77 \pm 0.39*	0.78 \pm 0.16*	1.04 \pm 0.15*	0.65 \pm 0.08*	0.38 \pm 0.10
Obese (12)	Low energy	5.30 \pm 0.41†	0.13 \pm 0.03†	4.63 \pm 0.44†	0.36 \pm 0.13†	0.54 \pm 0.07†	0.38 \pm 0.08†	0.17 \pm 0.02

*Significantly ($P, 0.05$) different from lean control-group dogs. †Significantly ($P, 0.05$) different from obese-group dogs fed a maintenance diet. VLDL-C = Very-low-density lipoprotein cholesterol. HDL-C = High-density lipoprotein cholesterol. LDL-C = Low-density lipoprotein cholesterol. VLDL-T = Very-low-density lipoprotein triglyceride. LDLHDL-T = LDL and HDL triglyceride.

higher (+72%) than of lean control-group dogs. The BW of obese-group dogs remained unchanged throughout the study (Table 1). The effect of sex on all measured values was tested for each group of dogs, but no significant effect was found.

Plasma ghrelin, leptin, glucose, and insulin concentrations—Compared with lean control-group dogs, obese-group dogs fed the complete and balanced diet had significantly lower plasma ghrelin (1.7-fold) and higher plasma leptin (5.8-fold) and insulin (1.9-fold) concentrations (Table 2). For obese-group dogs, the change to the low-energy diet resulted in a significant decrease in plasma leptin concentrations (–25%) and in a significant increase in plasma nonesterified fatty acid concentrations (+27%), whereas the plasma ghrelin, insulin, and glucose concentrations remained unchanged.

Plasma lipid and lipoprotein concentrations—Separation of lipoprotein fractions was confirmed by electrophoresis. Compared with lean control-group dogs, obese-group dogs fed the complete and balanced diet had significantly higher concentrations of cholesterol in total plasma (+41%) and in VLDL (+125%), HDL (+45%), and LDL (+58%) fractions and significantly higher concen-

trations of triglycerides in total plasma (+75%) and in the VLDL (+118%) fraction, whereas the increase in LDL-HDL triglyceride concentration (+28%) fractions was not significant (Table 3). For obese-group dogs, the change to the low-energy diet resulted in a significant decrease in cholesterol concentrations in total plasma (–16%) and in VLDL (–44%), HDL (–20%), and LDL (–53%) fractions and a significant decrease in triglyceride concentrations in total plasma (–48%) and in VLDL (–41%) and LDL-HDL (–56%) fractions.

Discussion

The primary objective of our study was to identify in dogs correlations among obesity and blood lipid and lipoprotein measurements commonly associated with obesity in humans. We chose to use chronically and grossly obese male and female dogs of various ages to mimic field conditions as closely as possible. However, mean values of age for obese and lean dogs in our study were similar to allow comparison between groups. We were interested in experimental conditions that were similar to naturally occurring obesity in dogs. Our dogs became obese by ad libitum consumption of a maintenance diet, and they were grossly obese with a stable BW for ≥ 1 year before the beginning of our study. Many

variables (diet, food intake, age, sex, and breed of dogs) were controlled to validate comparison between dogs. The effect of sex was not significant in our experiment. For lean and obese dogs fed either diet, blood values were modified in the same manner for male and female dogs. Therefore, data for male and female dogs were pooled. Results of our study indicate that chronic obesity in dogs is associated with significant increases in plasma lipid and lipoprotein concentrations (except for LDLHDL triglyceride concentration), significant increases in plasma leptin and insulin concentrations, and a significant decrease in plasma total ghrelin concentrations.

Long-term effects of hyperlipidemia in dogs are unknown, but hypercholesterolemia has been associated with ocular lesions²⁰ and hypertriglyceridemia may induce acute pancreatitis.²¹ Hyperlipidemia may also cause diabetes mellitus in humans.²² In contrast to humans, atherosclerosis is rare in dogs but has been observed in instances of hyperlipidemia secondary to hypothyroidism or diabetes mellitus or high-fat feeding with plasma cholesterol concentrations > 19 mmol/L.²³⁻²⁵ It was therefore of interest to evaluate the risk of high blood lipid concentrations in obese dogs. Blood cholesterol concentrations observed in obese dogs of our study were not compatible with atherogenic lesions. Moreover, HDL was the major (almost 90%) cholesterol carrier in obese and lean dogs. This profile is compatible with the resistance of dogs to spontaneous atheroma even in the obese state.²⁶ To our knowledge, this is the first time that a significant increase in most lipoprotein fractions has been reported. In a previous clinical study, Barrie et al²⁷ reported nonsignificant increases in cholesterol concentrations in lipoproteins in obese dogs. In another clinical study,²⁸ cholesterol, triglyceride concentrations in obese dogs were increased in serum and in all lipoprotein fractions except HDL₂, but the differences were significant only for total serum cholesterol and triglyceride concentrations and LDL triglyceride concentration. No information about the diet of dogs was available in these 2 studies.^{27,28} In a third study,⁷ obese insulin-resistant overfed experimental Beagles had higher total plasma triglyceride concentrations and VLDL and HDL triglyceride concentrations and lower HDL cholesterol concentrations, compared with healthy Beagles. From our results, it is difficult to determine whether increases in blood lipid concentrations were the result of obesity or high energy intake. Indeed, obese dogs consumed significantly more food than lean dogs, although their BW was stable.

In a previous experimental study,²⁹ however, total plasma cholesterol and triglyceride concentrations in obese dogs fed at lower intake amounts were similar to results of our study suggesting that obesity, rather than energy intake, was responsible for the modifications of blood lipid and lipoprotein measurements. Compared with the highest values for cholesterol and triglyceride found in the literature²⁶ for healthy pet dogs of multiple breeds, the concentrations in our study were within reference range except for the high HDL cholesterol concentration. In healthy dogs, the reference values are < 7.8 mmol/L for total plasma cholesterol concentra-

tion and < 1.7 mmol/L for total plasma triglyceride concentration (reference from commercial veterinary laboratory¹). Reference ranges determined by Jones and Manella³⁰ were 3.6 to 5.4 mmol/L for total plasma cholesterol concentrations and 0.6 ± 0.3 mmol/L for total plasma triglyceride concentrations. In lipoprotein fractions, cholesterol concentrations found in the literature for healthy pet dogs of multiple breeds were < 0.61 mmol/L for VLDL, 4.12 mmol/L for HDL, and 2.25 mmol/L for LDL. Triglyceride concentrations were < 2.2 mmol/L for VLDL and < 1.06 mmol/L for LDL-HDL.^{16,26,27,31} However, these values were from pet dogs and experimental dogs generally have lower values.³²

In our study, obese dogs had significantly higher plasma insulin and leptin concentrations, compared with lean dogs. The higher insulin concentrations could indicate a naturally occurring insulin resistance, although plasma glucose concentration measurements after withholding food were not higher, as observed by Kim et al.³³ In humans, insulin resistance and compensatory hyperinsulinemia promote enhanced production of triglycerides and cholesterol-rich lipoproteins by the liver, mainly VLDL triglyceride and LDL cholesterol.³⁴ High blood lipid concentrations observed in obese dogs of our study could therefore be secondary to naturally occurring insulin resistance.

High plasma leptin concentrations observed in obese dogs reflect a higher body fat mass.^{6,7} In obese dogs, insulin resistance could be related to visceral adiposity.³³ In our study, there was no attempt to differentiate visceral from subcutaneous adiposity.

Obese dogs in our study had low plasma ghrelin concentrations following the withholding of food, as observed in humans and rodents.^{11-13,35} Our results for obese dogs indicate that ghrelin is downregulated as a consequence of excess energy or excess energy storage.

Different nutritional approaches exist in the treatment of obese dogs. Among them, a low-fat high-fiber diet or a low-fat high-protein diet is usually proposed.³⁶ The second objective of our study was to determine how the differences in carbohydrate, protein, and fat content of a diet modify blood values in obese dogs. The diet change resulted in a spontaneous decrease in energy consumption by our obese dogs. The volume of food and the 90-minute time restriction may have limited the amount of food ingested. Consequently, effects of modification of diet composition cannot be separated from effects of a lower energy intake. Despite the significant decrease in energy intake, dogs had minimal weight loss. The length or the severity of energy restriction was probably not sufficient to result in a substantial weight loss. We found that the modification of diet composition and subsequent spontaneous energy restriction resulted in significant decreases in plasma lipid, lipoprotein, and plasma leptin concentrations and in a significant increase in plasma nonesterified fatty acid concentrations, whereas plasma ghrelin, insulin, and glucose concentrations remained unchanged.

Low fat intake could have induced a decrease in blood lipid concentrations, as observed in humans.^{37,38} Effects of a decrease in fat consumption on plasma lipoprotein concentrations have never been studied in obese dogs without weight loss. The influence of fat

content of diets has already been observed in working and pet Border Collies.³⁹ Two months of feeding a higher fat diet (20% vs 13% as fed) induced an increase in cholesterol concentrations in total plasma and in lipoproteins in working Border Collies. However, activity levels of dogs had induced some differences. Passive pet dogs had a higher VLDL triglyceride concentration when fed a diet with a high fat content. Results of another study⁴⁰ in Labrador Retrievers indicate that high-fat (20% to 25%) low-carbohydrate (26% to 33%) diets induce a significant increase in total plasma cholesterol concentration and in LDL cholesterol concentration, whereas a low-fat (13%) high-carbohydrate (44%) diet induces an increase in HDL triglyceride concentration. A third study⁴¹ showed that a high-fat moderately restricted protein diet resulted in increased total, free, and esterified cholesterol characterized by an increase in the alpha-fractions and in lecithin-cholesterol acyltransferase activity. In humans, low-fat diets without energy restriction have induced decreases in total plasma cholesterol concentration and in the LDL and HDL cholesterol concentrations but triglyceride concentrations were increased.^{42,43}

The influence of the protein content of the diet has already been studied in humans. High-protein low-carbohydrate diets without energy restriction in humans induce a decrease in total plasma triglyceride, VLDL triglyceride, total plasma cholesterol, and LDL cholesterol concentrations.^{44,45}

An effect of low energy intake alone is difficult to investigate in humans because a decrease in energy intake is often associated with modification of the diet composition and weight loss. In humans, Ræini-Sarjaz et al⁴³ found that an energy restricted diet with normal fat content improved blood lipid profiles by reducing triglyceride concentrations and the LDL-to-HDL ratio. However, the effect could have been the result of the low-carbohydrate content. In our study, a significant positive correlation was observed between energy intake and blood lipid concentrations.

In our study, feeding the low-energy diet resulted in a significant decrease in plasma leptin concentrations. Plasma leptin concentrations are more positively correlated with body fat content in dogs⁷ than with BW.⁶ Thus, the decrease in plasma leptin concentrations could reflect a decrease in body fat mass. This was confirmed by the significant increase in plasma nonesterified fatty acid concentrations, which suggests an increase in fat mobilization. A decrease in leptin concentrations is closely related to improvements in lipid metabolism.⁴⁶ In our study, a significant positive correlation was observed between leptin and blood lipid concentrations.

In humans, modification of the diet composition (low vs high glycemic index food) decreases body fat mass and improves blood lipid concentrations, before any weight loss.⁴⁷ Several hypotheses have been proposed for a decrease in body fat mass. These include an increase in fat or fat tissue oxidation, a decrease in oxidation of carbohydrate or muscle, an increase in lipolysis, and a decrease of lipoprotein lipase activity. In dogs, effects of diet composition on body fat content have been studied.³³ A slight increase (+8% on energy basis) in dietary fat without modification of energy

intake induces an increase in body fat mass, mainly abdominal fat, without a change in BW.³³ Insulin resistance is observed with an increase in adiposity.³³ Alteration in dietary intake and associated modifications of body composition could therefore result in metabolic variations. Modifications of blood lipid concentrations could be one of these metabolic variations.

However, leptin is also implicated in the acute regulation of food intake without modification of body composition.⁴⁸ A decrease in plasma leptin concentration and an increase in plasma nonesterified fatty acid concentrations could therefore also be the result of the negative energy balance. Decreased energy intake alone could result in the improvement of blood lipid and lipoprotein measurements. In our study, diet type had no effect on plasma ghrelin, insulin, or glucose concentrations in obese dogs.

Our results indicate that severe chronic obesity induces a significant increase in plasma cholesterol and triglyceride concentrations in dogs. Cholesterolemia and triglyceridemia were the result of an increase in lipid concentrations in all lipoprotein fractions. A significant increase in plasma insulin and leptin concentrations and a significant decrease in plasma ghrelin concentrations were also observed, as in humans, suggesting leptin and insulin resistance in obese dogs. A modification of the diet composition and a decrease in food intake resulted in beneficial effects on plasma lipid and leptin concentrations before any weight loss, whereas plasma ghrelin and insulin concentrations remained unchanged. These observations should be taken into account in the management of obese dogs, and blood lipid concentrations should be assayed. Modification of the diet composition and the resulting decrease in energy intake could improve blood lipid and lipoprotein measurements.

Appendix

Nutrient profiles as labeled of 2 diets fed to dogs of this study.

Nutrients	Diets	
	Maintenance	Low energy
Moisture (g/100 g)	8.0	8.0
Protein (g/100 g)	24.0	34.0
Fat (g/100 g)	16.1	9.5
Ash (g/100 g)	6.0	6.0
Starch (g/100 g)	38.0	14.8
TDF (g/100 g)	NL	27.0
Cellulose (g/100 g)	2.5	15.0
ME (kJ/g)	17.3	12.0

TDF = Total dietary fiber. ME = Metabolizable energy. NL = Not listed.

- Premium Croc Adult, Affinity- Petcare, Barcelona, Spain.
- Obesity Veterinary Diet, Royal Canin, Aimargues, France.
- Helena BioSciences Europe, Sunderland, UK.
- Technicon RA 1000, Bayer Diagnostics, Puteaux, France.
- Cholesterol Reagents, Technicon RA, Bayer Diagnostics, Puteaux, France.
- Triglycerides Reagents, Technicon RA, Bayer Diagnostics, Puteaux, France.
- Total ghrelin RIA kit, Linco Research, St Charles, Mo.
- INS-IRMA, Biosource Europe, Nivelles, Belgium.
- Free fatty acids half-micro test, Roche Diagnostics, Penzberg, Germany.
- Glucose Reagents, Technicon RA, Bayer Diagnostics, Puteaux, France.

- k. SAS Proc Mixed, SAS system release 8.2, SAS Institute Inc, Cary, NC.
 l. Laboratoire d'analyses vétérinaires J. Collard, Verviers, Belgium.

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