

Evaluation of the occurrence of metabolic syndrome in obese dogs and the role of caloric restriction diet as an adjuvant therapy

Avaliação da ocorrência de síndrome metabólica em cães obesos e o papel da dieta de restrição calórica como uma terapia adjuvante

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ABSTRACT: Metabolic syndrome, or metabolic dysfunction related to obesity in dogs, is a set of factors that may predispose comorbidities secondary to obesity. Adjuvant therapy with an energy-restricted diet, especially with low levels of carbohydrate and fat, is essential for weight loss, in addition to controlling snacks intake. The aim of this study was to evaluate the biochemical profile of obese dogs compared to lean dogs, and also to compare these profiles before and after a 30-day treatment, thus evaluating the possibility of obesity-related metabolic dysfunction and the action of adjuvant dietary therapy in this condition. Cholesterol and its fractions (HDL, LDL and VLDL), triglycerides, systemic blood pressure and glycemia of obese and lean dogs were measured, and seven obese dogs were treated only with a low-calorie diet with low levels of fat and carbohydrates for 30 days; these patients were evaluated before and after treatment. Obese dogs showed higher levels of triglycerides than lean dogs, and dogs treated with low-calorie diet presented weight loss and better outcomes related to biochemical profile, especially triglycerides levels, after treatment.

KEYWORDS: Obesity; dyslipidemia; hypertriglyceridemia.

RESUMO: A síndrome metabólica, ou disfunção metabólica relacionada à obesidade em cães, é um conjunto de fatores que podem predispor à obesidade. A terapia adjuvante com dieta com restrição calórica, especialmente com baixos níveis de carboidratos e gordura, é essencial para a perda de peso, além do controle da ingestão de petiscos. O objetivo do presente trabalho foi avaliar o perfil bioquímico de cães obesos em comparação aos de escora corporal normal e avaliar esses perfis em um grupo de animais obesos antes e após restrição calórica de 30 dias, verificando a ocorrência da disfunção metabólica relacionada à obesidade e o papel da dieta como tratamento adjuvante desta condição. Colesterol e suas frações (HDL, LDL e VLDL), triglicérides, pressão arterial sistêmica e glicemia de cães obesos e em escora corporal normal foram mensurados, e sete cães obesos foram tratados somente com uma dieta de baixa caloria com baixos níveis de gordura e carboidratos durante 30 dias; estes pacientes foram avaliados antes e após o tratamento. Cães obesos demonstraram valores séricos de triglicérides maiores quando comparados aos cães com escora corporal normal, e cães tratados com a dieta hipocalórica obtiveram perda de peso significativa e melhores resultados relacionados ao perfil bioquímico, especialmente dos níveis de triglicérides após o tratamento.

PALAVRAS-CHAVE: Obesidade; dislipidemia; hipertrigliceridemia.

INTRODUCTION

Metabolic syndrome was first described in 1923 in human beings, when Kylin reported a condition involving systemic arterial hypertension, hyperglycemia and hyperuricemia (increased levels of uric acid in the bloodstream) (PARIKH; MOHAN, 2012). In dogs, the term “metabolic syndrome” is still very controversial. Some authors use the term “obesity-related metabolic

dysfunction” (ORMD) instead of metabolic syndrome, and also suggest different parameters for characterization of the condition in dogs. In any case, obesity is considered the main triggering factor of this dysfunction (TVARIJONAVICIUTE et al., 2016; PIANTEDOSI et al., 2020). Currently, four parameters that occur in dogs are considered: systemic arterial hypertension, hyperlipidemia, insulin resistance and obesity (LUCENA et al., 2019).

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Obesity, which occurs when the weight of the animal exceeds 15% of its ideal weight, is considered the main triggering factor of ORMD in dogs (DALL'AGLIO et al., 2021). In humans, insulin resistance is considered the primary cause of metabolic syndrome, which in turn is correlated with abdominal obesity (TVARIJONAVICIUTE et al., 2019). Insulin resistance is characterized by decreased ability to metabolize blood glucose (LUCENA et al., 2019). Rahmouni et al. (2005) also state that insulin resistance has been shown to be a key factor for the development of the components of the syndrome. Hypertension and hyperlipidemia are also part of the components of the dysfunction. Systemic arterial hypertension consists of increased persistent systemic blood pressure above normal values, which can cause clinical or pathological sequelae (PIANTEDOSI et al., 2016).

Hypercholesterolemia and hypertriglyceridemia may occur by interaction between hereditary genetic factors with one or more other acquired factors. Excessive intake of lipids, lipogenesis or lipolysis contributes to hyperlipidemia (JERICÓ, 2015). Hypercholesterolemia has been associated with atherosclerotic diseases, pancreatitis and seizures in extreme cases (JEUSETTE et al., 2005; XENOULIS; STEINER, 2015).

Body fat percentage can be assessed by a subjective method, stratified into scales, which is called body condition score (BCS). The BCS is based on the inspection and palpation of the patient, with scales ranging from 1 to 9, as the patient on scale 1 is considered in cachexia, and the animal on a scale above 7 is considered obese. In patients with a score of 7, the ribs are hardly palpated, and there is no abundant layer of fat on the lumbar area, base of the tail, in addition to absence of waist; in animals with a score of 8, it is impossible to palpate the ribs by excess fat on it, or they are only palpable after hard pressure, in addition to large fat deposits on the lumbar area and base of the tail, abdominal distension and absence of waist; on the other hand, patients in score 9, considered morbidly obese, present attenuated fat deposits on the chest, spine, neck, limbs and tail base, in addition to evident abdominal distension (LAFLAMME, 1997).

Physical activity and diet are the main tools to treat and/or prevent metabolic syndrome (ZORAN, 2010). The treatment of obesity is based on a low-calorie and balanced diet, preferably with low levels of carbohydrates and fat, and higher levels of protein (FLANAGAN et al., 2017; CLINE et al., 2021).

The treatment and prevention of DMRO in animals and metabolic syndrome in humans are mainly based on the

premise of lifestyle change. Increased physical activity and diet characteristics are cornerstones to treat and prevent metabolic syndrome (ZORAN, 2010). Exercise helps in the prevention of metabolic diseases, considering that lack of physical activity is related to cardiovascular diseases and obesity. The effects of physical activity include: assisting in maintaining normal body weight, increasing insulin sensitivity, lowering systemic blood pressure, and raising HDL cholesterol levels, which reduce the risk of *diabetes mellitus* and heart disease (JERICÓ, 2015). The level of caloric restriction that should be adopted varies according to the degree of overweight. Weekly weight losses of 1% to 2% of the initial weight are considered satisfactory (FLANAGAN et al., 2017). The energy density of the diet is reduced by decreasing levels of fat and increasing levels of fiber and protein (WEBER et al., 2007), maintaining adequate levels of essential nutrients such as amino acids, vitamins and minerals (FLANAGAN et al., 2017).

The present study aimed to evaluate the biochemical profile (cholesterol and its fractions, triglycerides, glycemia) and systemic blood pressure of obese dogs compared to lean dogs, and also to verify these same parameters before and after dietary therapy with caloric restriction for 30 days, thus evaluating the occurrence of obesity-related metabolic dysfunction and the role of diet as adjuvant therapy of this condition.

MATERIALS AND METHODS

Nineteen dogs were selected, without any restriction related to gender and breed, which were divided into three groups: group 1 consisting of six healthy animals with a body score of 4/5 (control group), group 2 that included six obese animals with body score between 8 and 9, who were not subsequently treated, and group 3 consisting of seven obese dogs (BCS 9/9), considered to be morbidly obese, who have undergone adjuvant dietary therapy with caloric restriction (Ração Farmina Obesity®) during 30 days. The owners responsible for the dogs signed a consent form for participation in the study, in which they compromised to offer only the prescribed food in the quantities calculated during the treatment period, without any other type of food or snack. The daily amount was divided into three meals, which were offered at times determined by the owners according to their habits. The daily amount of food was calculated by the formula: factor x (ideal weight)^{0.75}, the ideal weight considered 5% less than the current live weight. The factors used in the formula are shown in Figure 1.

Fat percentage (%)	25-35%		35-45%		>45%	
BCS	7		8		9	
Daily energetic intake (factor)	Male	Female	Male	Female	Male	Female
	85	80	75	65	60	55

Figure 1. Factors used to calculate caloric restriction diet based on body score and animal gender (APTEKMANN, 2009)

Animals with previously diagnosed comorbidities, such as neoplasms, heart diseases, endocrinopathies, were not included in the experiment.

The 19 dogs were evaluated and the body condition score (BCS) was confirmed by direct inspection and palpation, based on the information described by Laflamme (1997). Animals with BCS of 8 or 9 were considered obese. Animals with ideal body condition were those with BCS 4 or 5, with palpable ribs without excessive fat coverage, without abdominal distention, easily visualized waist and apparent abdominal recess.

The biochemical profile (glycemia and serum levels of triglycerides, cholesterol and fractions [LDL, HDL and VLDL]) of all patients was evaluated, in addition to the measurement of systolic blood pressure by doppler method.

Systolic blood pressure was measured by the Doppler¹ method before blood collection, by obtaining the average of seven consecutive measurements, which were performed in a calm environment, after 30 minutes of arrival, seeking to minimize possible changes caused by stress. The animals were kept only by physical restraint in right lateral decubitus for blood collection. Four milliliters of blood were collected from patients who were fasting for 12 hours, without water restriction, by jugular venopuncture, with prior trichotomy and antisepsis of the collection site. Blood was inserted in tubes without anticoagulant to obtain blood serum after centrifugation (10 minutes) (ROMÃO et al., 2013). During blood collection, a drop of whole blood was used from each dog to measure glycemia, performed by glucometer². The tubes were taken to the Clinical Laboratory of the Veterinary Hospital of Faculdades Integradas de Ourinhos for analysis. Blood samples No laboratório, as

amostras de sangue were centrifuged at 3000 rpm³ to obtain the serum, which was used for serum measurements of triglycerides, and cholesterol and its fractions of each animal. For these analyses, commercial kits⁴ were used following the methodologies recommended by the manufacturer.

The reading and confirmation of the results were evaluated by a semi-automatic biochemical analyzer⁵. The reference values used followed the hematology guide of Weiss and Wardrop (2010).

A comparison was made between the evaluations of group 1 (lean animals) and group 2 (obese animals without treatment), and another research comparing results of group 3 (obese animals submitted to diet) at the beginning (M0), and end (M1) of the treatment.

The results were statistically analyzed. In the comparison between groups 1 and 2 (lean x obese), the Kolmogorov-Smirnov normality test was used. The variables that did not pass the normality test were presented as median (min-max) and compared by the Mann-Whitney test; on the other hand, the variables that passed the normality test are presented as mean \pm standard deviation, and were compared by unpaired t-test. For the comparison between the beginning and end of the treatment of group 3, the Shapiro-Wilk normality test was performed.

The study was approved by the ethics committee of the use of animals (CEUA) of the Faculdades Integradas de Ourinhos, under protocol number 2016/022.

RESULTS

In the comparison between groups 1 and 2 (Tables 1 and 2) in relation to the values of systolic blood pressure, glycemia, cholesterol and its fractions, and triglycerides, the

Table 1. Comparison of systolic blood pressure, glycemia, cholesterol and fractions, and triglycerides values among lean and obese dogs.

Groups	Gender	Neutered	SBP (mmHg)	Biochemical variables (mg/dL)					
				Glycemia	Cholesterol	Triglycerides	HDL	LDL	VLDL
Lean	M	No	140	98	89.0	479	24.5	55.0	9.6
	M	Yes	120	73	109.2	28.0	38.6	65.0	5.6
	F	No	120	92	201.5	58.4	23.0	166.9	11.6
	M	Yes	150	88	229.9	133.3	69.2	134.1	26.6
	F	Yes	120	92	141.1	63.9	36.9	91.5	12.7
	F	No	110	82	237.9	91.0	35.0	184.7	18.2
Obese	F	No	120	87	206.4	115.0	44.5	138.9	23.0
	F	No	150	106	362.0	95.5	67.0	276.1	18.9
	M	Yes	150	92	186.9	108.0	37.0	128.3	21.6
	M	Yes	150	96	309.8	130.7	60.8	222.9	26.1
	F	Yes	120	76	281.9	322.9	61.5	155.8	64.6
	F	Yes	120	74	246.0	375.2	54.0	117.0	75.0

¹ Doppler

² G-Tech Free 1[®], Glicomed

³ Centrifuge CELM Combate[®], LS3 Plus, Barueri - SP

⁴ Labtest[®] - Labtest Diagnóstica S/A, Lagoa Santa-MG

⁵ Bioplus BIO-2000 semi-automatic biochemical analyzer

latter values were significantly higher in the group of obese animals. Half (3/6) of obese animals had hypercholesterolemia (> 275 mg/dL), while in the control group this was not observed. Two animals in group 2 showed increased LDL fraction levels, and all (6/6) of obese animals presented values of the VLDL fraction above normal levels. Obese animals presented higher cholesterol, triglyceride, LDL and VLDL values than lean patients. One animal presented arterial hypertension (≥ 160 mmHg), and the values of group 2 (obese) were higher than the values of the animals with normal body score. No animal presented fasting hyperglycemia, and in the comparison of the values there was no significant difference between lean and obese.

In the comparison between groups 1 and 2, the frequency of neutered animals was higher in the group of obese animals (66.66%) than in lean animals (50%) (Table 1). In a total

of six obese animals in group 2, four of them (4/6; 66.66%) were females.

In the comparison between obese animals in group 3 (before and after dietary therapy with caloric restriction [Tables 3 and 4]), a significant decrease was observed after the diet of cholesterol, LDL, VLDL, blood pressure, glycemia and mainly triglyceride values. After treatment, group 3 showed decreased levels of total cholesterol, LDL and VLDL fractions, and triglycerides. There was also an increase in the HDL fraction. All animals lost weight, with mean baseline values of 8.2 kg (2.8 - 40.8), and 7.7 kg (2.7 - 36.8) after treatment. Regarding systolic blood pressure, only one obese animal in group 3 presented hypertension before and after the diet (160 mmHg); however, in four animals (57%) there was a decrease in the systolic blood pressure. In the independence sample T-test, there was a significant correlation between obesity and serum triglyceride levels.

Table 2. Comparison of systolic blood pressure, glycemia, cholesterol and its fractions, and triglycerides between lean and obese dogs. (* $p < 0.05$)

Variable	Group		P
	Lean	Obese	
SBP (mmHg)	120 (110 - 150)	135 (120 - 150)	0.4156
Glycemia (mg/dL)	87.5 \pm 8.8	88.5 \pm 12.2	0.8741
Cholesterol (mg/dL)	168.1 \pm 63.7	265.5 \pm 65.7	0.0261*
Triglycerides (mg/dL)	61.1 (28.0 - 133.3)	122.8 (95.5 - 375.2)	0.0260*
HDL (mg/dL)	37.9 \pm 16.7	54.1 \pm 11.4	0.0769
LDL (mg/dL)	116.2 \pm 54.0	173.2 \pm 62.8	0.1228
VLDL (mg/dL)	12.2 (5.6 - 26.6)	24.6 (18.9 - 75)	0.0260*

Table 3. Comparison of systolic blood pressure, glycemia, cholesterol, triglycerides, HDL, LDL and VLDL fractions, at the beginning (day 0) and end of treatment (day 30) in obese animals before and after treatment with adjuvant diet.

Animal	Gender	Neutered	Day	Weight (kg)	SBP (mmHg)	Biochemical variables (mg/dL)					
						Glycemia	Cholesterol	Triglycerides	HDL	LDL	VLDL
1	M	Yes	0	11.2	130	76.0	163.3	112.7	112.2	28.6	22.5
			30	10.9	100	75.0	135.8	45.6	120.7	6.0	9.1
2	F	Yes	0	6.4	130	86.0	295.9	89.5	74.0	204.0	17.9
			30	6.1	120	86.0	186.0	59.8	131.0	43.1	11.9
3	F	Yes	0	2.8	130	97.0	155.7	34.0	46.0	102.9	6.8
			30	2.7	130	89.0	135.1	32.5	46.2	82.4	6.5
4	F	No	0	3.8	140	76.0	229.0	123.6	66.0	138.3	24.7
			30	3.4	130	76.0	106.8	57.7	78.8	16.5	11.5
5	F	Yes	0	9.0	130	97.0	224.2	117.4	73.5	127.3	23.4
			30	8.7	120	94.0	153.4	109.0	77.8	53.8	21.8
6	F	Yes	0	40.8	160	81.0	190.0	212.3	106.3	41.2	42.5
			30	36.8	160	74.0	162.5	56.9	115.8	35.4	11.3
7	M	Yes	0	8.2	110	87.0	226.6	124.7	46.2	155.5	24.9
			30	7.7	110	85.0	120.8	85.7	73.9	29.8	17.1

Tabela 4. Comparison of systolic blood pressure, glycemia, cholesterol and its fractions, and triglycerides values in obese animals before and after treatment with adjuvant diet. (*p < .05)

Variable	Moment		p
	Day 0 (M0)	Day 30 (M1)	
Weight (kg)	8.2 (2.8 – 40.8)	77 (2.7 – 36.8)	0.0156*
SBP (mmHg)	133 ± 15	124 ± 19	0.0781
Glycemia (mg/dL)	85.7 ± 8.8	82.7 ± 7.8	0.0511
Cholesterol (mg/dL)	212.1 ± 47.8	142.9 ± 26.6	0.0060*
Triglycerides (mg/dL)	116.3 ± 53.0	63.9 ± 25.6	0.0372*
HDL (mg/dL)	74.9 ± 26.2	92.0 ± 30.9	0.0599
LDL (mg/dL)	114.0 ± 62.3	38.1 ± 25.2	0.0171*
VLDL (mg/dL)	23.2 ± 10.6	12.7 ± 5.1	0.0374*

DISCUSSION

Tvarijonaviciute et al. (2012) proposed the term “obesity-related metabolic dysfunction” (ORMD) and suggested different parameters for characterizing this condition in dogs. Based on these parameters, in a total of 13 obese animals tested in this study, only one obese animal would be classified as a case of ORMD.

Castration is also included as a risk factor for obesity in dogs due to decreased metabolism, increased appetite and replacement of muscle mass by adipose tissue due to a lower concentration of androgenic hormones (GERMAN, 2006). In the present study, the group of obese animals had more neutered animals (66.66%) than the group of lean animals (50%), corroborating results that described a prevalence of 67% of neutered animals on the obese population (JERICÓ et al., 2006). In the present study, in a total of six obese animals in group 2, four of them (66.66%) were females. This sexual predisposition occurs due to the lower concentration of androgen hormones and the lower metabolic rate in females (MCGREEVY et al., 2005; PORSANI et al., 2020). The small number of animals did not allow the observation of statistical difference between males and females. However, the literature states that castrated males have a tendency to obesity similar to females and, therefore, that the reproductive status is a factor that influences the occurrence of obesity, but without influence of gender (EDNEY; SMITH, 1986; MAO et al., 2013; PORSANI et al., 2020).

The levels of serum total cholesterol and its fractions (HDL, LDL and VLDL) and triglycerides were significantly higher in the group of obese animals, corroborating a study that verified all these variables measured higher in the obese group compared to the control group (TRIBUDDHARATANA, et al 2011). Thus, it is assumed that chronic obesity in dogs directly influences hyperlipidemia (JEUSSETE et al. 2005; YAMKA et al. 2006), resulting in an important frequency of obese animals with hypertriglyceridemia and hypercholesterolemia

(BRUNETTO 2011). Obese dogs have increased levels of triglycerides and cholesterol when compared to dogs with adequate body condition, and tend to have a significant reduction in these values after the establishment of a low-calorie diet (low energy intake and high protein levels) (JEUSSETTE et al., 2005; YAMKA et al., 2006; TRIBUDDHARATANA et al., 2011). High serum levels of triglycerides, free fatty acids (AGL), beta-hydroxybutyrate and cholesterol were observed, induced by high-fat diets (THIES et al., 2003).

Thus, the use of low-calorie diets with high levels of dietary fibers and reduced lipid contents allows weight loss and reduction of hyperlipidemia (FLANAGAN et al., 2017). It can be observed a decrease in the total cholesterol, LDL, VLDL, and especially in triglyceride levels in obese dogs of group 3 submitted to caloric restriction for 30 days. Triglyceride levels decreased significantly, which can also be observed in obese dogs that underwent diet change and weight loss that showed improvement in hyperlipidemia; triglyceride levels decreased more significantly than cholesterol levels after the onset of a low-calorie diet; this was also observed in 12 obese dogs that underwent a low energy and high protein diet over a period of 30 days, when a decrease in cholesterol, VLDL and LDL levels were observed, and a significantly greater decrease in triglycerides levels (JEUSSETE et al, 2005). This can be explained by the fact that high-carbohydrate diets are associated with increased plasma triglyceride concentrations, HDL reduction and insulin resistance.

In the diet, several factors influence hyperlipidemia, including the amount of carbohydrates, the type of sugar (glucose, fructose, sucrose, lactose), the nature of starch and the size of the carbohydrate particle (RICCARDI; RIVELLESE, 2000). A calorie-restricted diet in the treatment of obesity significantly reduces the serum triglycerides levels, as dietary carbohydrate intake and obesity are associated with high levels of triglycerides (YAMADA et al., 2021). The use of low-calorie diets with high protein and fiber levels and low carbohydrate and

lipid levels reduces glycemia, cholesterol and triglyceride levels (CLINE et al., 2021), which proves that diets with low lipid values allow the reduction of hyperlipidemia and decrease in these values even in those animals that show cholesterol, triglycerides, glycemia and blood pressure parameters within the normal range (PEREIRA NETO et al., 2010; VERKEST, 2014; PÉREZ-SÁNCHEZ et al., 2015). For this, the overweight animal should not receive any snack or food other than its permitted feed (CLINE et al., 2021).

Carbohydrate-rich diets are associated with higher postprandial glycemia values (PIATENDOSI et al., 2020). Rand et al. (2003) associated a diet with high levels of carbohydrates with higher postprandial glycemia peak values. Even though hyperglycemia was not verified, the reduction in serum glucose values was observed in 10 obese dogs after weight loss in another study (ZOTELLI et al., 2014), which also occurs in the present study, in which, even though there was no fasting hyperglycemia detected, glycemia levels decreased in 71.42% of obese animals in group 3 after diet with low carbohydrate levels.

An obese animal in group 3 presented arterial hypertension (≥ 160 mmHg), which is in line with a study where arterial hypertension was observed in 45% of the 69 obese animals tested (JERICÓ et al., 2006). The statistical difference in values can be explained by the difference in the number of animals tested. Hypertension in obese animals is caused by several factors, including hyperleptinemia, activation of sympathetic nervous system (SNS) and renin-angiotensin-aldosterone system (RAAS), and the physical compression of

the kidneys itself by visceral adipose tissue, although these are factors most studied in humans (RAHMOUNI et al., 2005; DA SILVA et al., 2020). In 57% of obese animals in group 3, there was a decrease in blood pressure values, corroborating results that show the reduction of blood pressure values after diet of all obese animals tested (ZOTELLI et al., 2014). The decrease in blood pressure after weight loss is explained by the decrease in visceral fat, which is responsible for activation of RAAS through leptin twice as high when compared to subcutaneous adipose tissue (ENGELI et al., 2005; DA SILVA et al., 2020). The decrease in blood pressure values is one of the benefits of weight loss, even though these values are already within normal limits (PEREIRA NETO et al., 2014; VERKEST, 2014; ZOTELLI et al., 2014; PÉREZ-SÁNCHEZ et al., 2015). Thus, the hypocaloric diet is shown to be important in reducing visceral adipose tissue, thus reducing blood pressure (FLANAGAN et al., 2017).

CONCLUSION

The levels of cholesterol and some of its fractions (LDL and VLDL), triglycerides and systemic blood pressure were significantly higher in obese dogs, and females were more affected by obesity than males. The results of the study show that the use of a hypocaloric diet allows weight loss and improvement of these parameters results over a 30-day period, of which cholesterol, triglycerides, blood glucose and blood pressure can be cited. It is important to maintain optimal body weight in order to avoid compromising normal physiological functions and metabolic disorders caused by obesity, such as obesity-related metabolic dysfunction.

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