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ORIGINAL ARTICLE

Copaiba oleoresin presents anti-obesogenic effect and mitigates inflammation and redox imbalance in adipose tissue

Luciana Ortega TELLES[1], Bianca Sulzbacher da SILVA[1], Angélica Macedo Borges PAULINO[1], Sabrina Trigueiro MENDONÇA[1], Valéria Dornelles Gindri SINHORIN[2], Milena Campelo Freitas de LIMA[3], Valdir Florêncio VEIGA JUNIOR[4], Carla Regina ANDRIGHETTI[1], André Ferreira do NASCIMENTO[1], Gisele Facholi BOMFIM[1], Renata Azevedo Melo LUVIZOTTO[1*]

[1] Universidade Federal de Mato Grosso (UFMT), Instituto Ciências da Saúde, Sinop, MT, Brazil

[2] Universidade Federal de Mato Grosso (UFMT), Instituto de Ciências Naturais, Humanas e Sociais, Sinop, MT, Brazil

[3] Universidade Federal do Amazonas (UFAM), Departamento de Química, Manaus, AM, Brazil

[4] Instituto Militar de Engenharia, Departamento de Engenharia Química, Rio de Janeiro, RJ, Brazil

* Corresponding author: renata.nascimento@ufmt.br; https://orcid.org/0000-0001-9548-6338

ABSTRACT

Obesogenic diets lead to fat accumulation and dysfunctional adipose tissue. Active compounds were described in copaiba oleoresin, which presents anti-inflammatory, antimicrobial, and antioxidant properties. However, there are no data regarding the effects of copaiba oleoresin in adipose tissue. Therefore, we tested the hypothesis that the copaiba oleoresin could prevent or minimize obesity and adipose tissue inflammation and oxidative stress in response to a high sucrose diet. Male *Wistar* rats were randomly assigned to receiving commercial chow (C, n = 8), commercial chow and 30% sucrose added to the drinking water (S, n = 8), or commercial chow and 30% sucrose added to the drinking water + copaiba oleoresin (S+CO, n = 8). Copaiba oleoresin was given at a dose of 200 mg kg^{-1} day^{-1} by gavage for eight weeks. C and S animals received vehicle, at equivalent volume, by gavage. At the end of the experiment, blood samples and epididymal adipose tissue were collected for biochemical, inflammatory, and oxidative stress analyses. Copaiba oleoresin supplementation prevented weight gain, adiposity, insulin resistance, and increased IL-1β levels. Additionally, copaiba oleoresin partially attenuated the increase in fasting glucose levels, lipids, and IL-6 levels, and improved the redox status in adipose tissue. Our results suggest that the use of copaiba oleoresin could be a good strategy for prevention of obesity and its complications.

KEYWORDS: natural products; high sucrose diet; obesity; *Copaifera*

Óleo-resina de copaíba apresenta efeito antiobesogênico e atenua inflamação e desequilíbrio redox no tecido adiposo

RESUMO

Obesidade leva ao acúmulo de gordura e disfunção do tecido adiposo. Compostos ativos foram descritos no óleo-resina de copaíba, que apresenta efeitos anti-inflamatórios, antimicrobiais e antioxidantes. Contudo, não há dados sobre os efeitos do óleo-resina de copaíba sobre o tecido adiposo. Assim, testamos a hipótese de que o óleo-resina de copaíba pode prevenir ou minimizar a obesidade, e inflamação e estresse oxidativo no tecido adiposo em resposta à dieta rica em sacarose. Ratos *Wistar* machos foram casualmente divididos para receber dieta padrão (C, n = 8), dieta padrão e 30% de sacarose na água de beber (S, n = 8), e dieta padrão e 30% de sacarose na água de beber + óleo-resina de copaíba (S+CO, n = 8). O óleo-resina de copaíba foi administrado via gavagem (200 mg kg^{-1} dia^{-1}), por oito semanas. Animais dos grupos não suplementados (C e S) receberam veículo via gavagem, em volume equivalente ao oferecido ao grupo S+CO. Ao final do experimento, amostras de sangue e tecido adiposo epididimal foram coletadas para análises bioquímicas, inflamatórias e de estresse oxidativo. A suplementação com óleo-resina preveniu ganho de peso, adiposidade, resistência à insulina e aumento de IL-1β, inibiu parcialmente o aumento nos níveis de glicemia de jejum, lipídeos e níveis de IL-6 no tecido adiposo, além de melhorar o sistema de defesa antioxidante no tecido. Nossos resultados sugerem que o óleo-resina de copaíba pode ser usado como uma boa estratégia para a prevenção da obesidade e suas complicações.

PALAVRAS-CHAVE: produtos naturais; dieta rica em sacarose; obesidade; *Copaifera*

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INTRODUCTION

Obesogenic diets, consisting of energy-dense foods that are generally high-fat and/or high-sugar, lead to fat mass accumulation (Romieu *et al*. 2017). Adipose tissue is known to perform a number of functions, including storage of excess energy, through its hypertrophy capacity. However, adipocytes possess a limited capacity to store fat, and when their maximal growth is reached, they become more lipolytic and present altered adipokines secretion, which is associated with increased inflammation and oxidative stress in adipose tissue (Rogero and Calder 2018; Longo *et al*.2019).

It is currently well established that adipose tissue is a dynamic endocrine organ, able to release a large variety of proteins, which are biologically active and participate in physiological and pathological processes through endocrine, paracrine, and autocrine signaling pathways (Longo *et al*. 2019). Given that adipose tissue is an important metabolic organ, it is necessary to identify new compounds that could be used as dietary functional ingredients as a therapeutic approach against adipose tissue dysfunction and related comorbidities.

Recent studies have shown that medicinal plants or plant-based medicinal products have beneficial effects on obesity and related complications (Zielinska-Blizniewska *et al*. 2019; Telles *et al*. 2020; Von Dentz *et al*. 2020). Copaiba oleoresin, a product from the copaiba (*Copaifera* sp.) tree, is one of the most important renewable natural remedies for the indigenous people from the Amazon region and its use is widely diffused due to its various pharmacological properties, particularly its anti-inflammatory activity. (Pieri *et al*. 2009; Vásques *et al*. 2014; Luca *et al*. 2018). The oleoresin has resinous acids and volatile compounds, being composed of a high concentration of sesquiterpenes, as well as a small concentration of diterpenes (Arruda *et al*. 2019). Copaiba oleoresin has been shown to have, among others, anti-inflammatory and antioxidant properties (Ames-Sibin *et al*. 2019; Telles *et al*. 2020).

Copaiba oleoresin supplementation prevented obesity development and was associated to the hepatic redox system and steatosis improvement (Telles *et al*. 2020). However, to the best of our knowledge, no study has investigated the effect of copaiba oleoresin on adipose tissue in a high sucrose diet model. Therefore, the present study tested the hypothesis that the copaiba oleoresin could prevent or minimize obesity and adipose tissue inflammation and oxidative stress in response to a high sucrose diet.

MATERIAL AND METHODS

Copaiba oleoresin acquisition and compound identification

Copaiba oleoresin was commercially acquired at Fazenda São Nicolau, located at Cotriguaçu municipality, northwest of Mato Grosso state, Brazil (09°49'09.0"S, 58°15'31.1"W).

High-resolution gas chromatography-mass spectrometry (HRGC-MS) analyses of the copaiba oleoresin was performed by using a Thermo Scientific model Trace GC Ultra instrument following the methodology described by Barbosa *et al*. (2013). The sample was esterified with trimethylsilyldiazomethane (TMSD) according to the method described by Migowska *et al*. (2010). The chemical constituents were identified by comparing their retention indices (RI) with the literature, by co-injection of commercially available β-caryophyllene and α-humulene, and by comparing the fragmentation profiles obtained with the Mass Spectral Library Wiley, 8th edition. The percentage of each constituent was expressed in relative concentration values (%). The diterpene analysis was performed using previously isolated and identified substances (Veiga-Junior *et al*. 2006).

Animals and experimental protocol

Male Wistar rats weighing approximately 250 g were obtained from the breeding facilities at Universidade Federal de Mato Grosso, UFMT. The animals were randomly assigned to three treatments, to receive (1) commercial rat chow (C, n = 8); (2) commercial chow and 30% sucrose added to the drinking water (S, n = 8); or (3) commercial chow and 30% sucrose added to the drinking water + copaiba oleoresin (S+CO, n = 8). All animals had free access to water and the commercial chow (Nuvilab® Cr-1, Nuvital, Colombo, Paraná, Brazil). The S+CO group received copaiba oleoresin at a dose of 200 mg kg^{-1} day^{-1} (Gonçalves *et al*. 2014) by gavage for eight weeks, while the C and S groups were administered the same volume of vehicle every morning. The rats were housed in groups (4 rats per cage) at the Cardiovascular and Metabolic Diseases Laboratory of UFMT Sinop campus, under controlled temperature (22–26 °C) and lighting (12 h light-dark cycle). Food and water intake were measured daily, and body weight was assessed weekly. The animals were sacrificed by decapitation under deep sodium thiopental anesthesia (intraperitoneal (i.p.) injection; 50 mg kg^{-1}). Serum and epididymal adipose tissue were collected and stored at –80 °C until required for analysis. The experiment followed the principles of the National Institute of Health Guide for the Care and Use of Laboratory Animals (NIH Publications No. 80-23) revised 1996. The study protocol was approved by the local Committee for Ethics in Animal Research (CEUA/UFMT protocol # 23108.050625/2019-38).

Nutritional and body fat analysis

The nutritional composition of the commercial chow is presented in Table 1. The caloric content values of the chow and the sucrose solution were used to obtain the total caloric intake. The adipose tissue depots were isolated, and epididymal, retroperitoneal, and mesenteric adipose tissues were weighed. The sum of these depot masses was considered as the visceral fat, and was used to calculate the adiposity index (Nascimento *et al*. 2011) to confirm obesity in the animals.

Table 1. Nutritional composition of the diet fed to the rats.

Components	Commercial chow Nuvilab® Cr-1
Carbohydrate (g kg^{-1})	655
Protein (g kg^{-1})	220
Fat (g kg^{-1})	40
Fibers (g kg^{-1})	40
Vitamin mixture (g kg^{-1})	10
Mineral mixture (g kg^{-1})	35
Total (g kg^{-1})	1000
Energy (kcal g^{-1})	3.8

Biochemical serum analysis

The serum was used for the spectrophotometric analysis of glucose (Analisa®, Cat# 434, Belo Horizonte, MG, Brazil), total cholesterol (Analisa®, Cat# 460, Belo Horizonte, MG, Brazil), high-density lipoprotein cholesterol (HDL) (Analisa®, Cat# 413, Belo Horizonte, MG, Brazil) and triglycerides (TG) (Analisa®, Cat# 459, Belo Horizonte, MG, Brazil). Very low-density lipoprotein cholesterol (VLDL) was determined according to the formula: VLDL (mg dL^{-1}) = TG (mg dL^{-1})/ 5. Low-density lipoprotein cholesterol (LDL) was determined by subtracting the HDL and the VLDL values from the total cholesterol values (Friedewald *et al.* 1972).

Adipokine levels

The concentration of IL-6 (R&D Systems, Cat# R6000B, Minneapolis, MN, USA), IL-1β (R&D Systems, Cat# RLB00, Minneapolis, MN, USA), IL-10 (R&D Systems, Cat# R1000, Minneapolis, MN, USA) was analyzed in the epididymal adipose tissue. We homogenized 300 mg of frozen epidydimal adipose tissue with 1 mL of ice-cold phosphate-buffer saline (pH 7.4). The homogenate was centrifuged at 10,000 rpm for 15 min. The medium layer was collected and analyzed using commercial ELISA kits according to the manufacturer's instructions.

Redox status analysis

Redox status was evaluated in homogenates of epidydimal adipose tissue samples. The frozen epidydimal adipose tissue was homogenized with 10 volumes of ice-cold 0.1 M potassium phosphate buffer (pH 7.4). The homogenate was centrifuged at 10,000 rpm for 15 min. The medium layer was collected and used to perform the analysis. Oxidative damage was assessed by the determination of thiobarbituric acid reactive substances (TBARS), according to the method described by Buege and Aust (1978), and carbonylated protein assay, using a method adapted from Mesquita *et al.* (2014). Briefly, 100 µL of homogeneous to 100 µL 2,4-dinitrophenylhydrazine (DNPH) (10 mM in 2 M HCl) was used. The samples were incubated for 10 minutes at room temperature and then 50 µL of NaOH (6 M) were added and incubated again for 10 minutes at room temperature. The reading was performed at 450 nm. Enzymatic and nonenzymatic antioxidants were evaluated through the activity of superoxide dismutase (SOD), measured according to Misra and Fridovich (1972), glutathione S-transferase (GST), determined according to Habig *et al.* (1974), reduced glutathione (GSH), determined according to Sedlack and Lindsay (1968), and ascorbic acid (Vit C) levels, determined according to Roe (1954).

Statistical analysis

All response variables were normally distributed and complied with variance homogeneity (according to Kolmogorov-Smirnov tests and Levene tests, respectively) The comparison among treatments was made using parametric one-way analysis of variance (ANOVA), followed by Student Newman Keuls' *post hoc* test. All statistical analyses were performed using Sigma Stat, version 3.5 for Windows (Systat Software, Inc.), and a P value < 0.05 was considered statistically significant.

RESULTS

Chemical composition of the copaiba oleoresin

The copaiba oleoresin mainly contained sesquiterpenes such as *trans*-α-bergamotene (20.24%), β-selinene (11.48%), β-bisabolene (8.80%) and α-copaene (8.26%), and diterpenes, mainly kaurenoic acid (7.80%) (Table 2).

Biological parameters

At the end of the experimental period, S rats showed higher caloric intake and body weight, as well as increased body weight gain and mesenteric and retroperitoneal adipose tissue deposits, reflecting in higher visceral adipose tissue and adiposity index, when compared to the C group (Table 3). Copaiba oleoresin supplementation did not affect caloric intake, but prevented weight gain and fat deposition, as these variables did not differ significantly from the control group.

The high sucrose diet, alone or in combination with copaiba oleoresin supplementation, did not alter HDL levels (Table 4). The levels of total cholesterol, LDL, TG, VLDL, TG/HDL ratio and fasting glucose were significantly higher in the S group compared with the C group, but did not differ significantly from the control in the S+CO group.

Effect on inflammatory adipokines

Regarding IL-10 levels, no statistical difference was observed among groups. The IL-6 and IL-1β levels in epididymal adipose tissue were significantly higher in the S group compared to the C group, but did not differ significantly from the control in the S+CO group. The effect of copaiba oleoresin supplementation partially attenuated IL-6 levels, and efficiently reduced IL-1β levels, restoring them to C group levels (Figure 1).

Effect on adipose tissue redox status

GST, Vit C, and carbonylated protein did not vary significantly among treatments. Antioxidant defenses (SOD

Table 2. Chemical composition of copaiba (*Copaifera* sp.) oleoresin from Cotriguaçu, Mato Grosso state, Brazil, determined by HRGC-MS analyses.

Compound	Retention index	Concentration (%)
α-cubebene	1348	0.49
Cyclosativene	1369	0.28
α-copaene	**1374**	**8.26**
β-cubebene	1387	2.02
β-elemene	1389	4.10
Cyperene	1398	0.24
β-caryophyllene	1417	0.49
***trans*-α-bergamotene**	**1432**	**20.24**
cis-β-farnesene	1440	1.27
epi-β-santalene	1445	0.29
α-humulene	1452	0.09
trans-β-farnesene	1454	1.09
allo-aromadendrene	1458	0.61
4,5-di-*epi*-aristolochene	1471	0.13
γ-muurolene	1478	0.13
Germacrene D	1480	1.86
β-selinene	**1489**	**11.48**
α-selinene	1498	5.69
cis-α-bisabolene	1506	1.17
β-bisabolene	**1505**	**8.80**
7-*epi*-α-selinene	1520	0.38
δ-cadinene	1522	2.06
Junenol	1618	0.19
Selin-11-en-4-α-ol	1658	1.08
epi-α-bisabolol	1683	0.12
β-copaen-4-α-ol	1590	0.39
Kaur-16-en acid methyl ester	-	0.45
Kaurenoic acid methyl ester	**-**	**7.80**
Copalic acid methyl ester	-	1.78
Daniellic acid methyl ester	-	0.58
Identified constituents (%)		**83.55**
Sesquiterpene content (%)		**72.94**
Diterpenes (%)		**10.60**

and GSH) were significantly lower, and TBARS were significantly higher, in the S group compared to the C and S+CO groups (Figure 2).

DISCUSSION

In this study, copaiba oleoresin was effective in preventing weight gain, high adiposity, and insulin resistance, displaying an antiobesogenic effect in a rodent model. Copaiba oleoresin was also associated to improved adipose tissue inflammation and redox status. These results support our hypothesis regarding the beneficial effects of copaiba oleoresin on adipose tissue in a high sucrose diet model.

Table 3. Caloric intake and morphological characteristics of rats subjected to three treatments for eight weeks. C: control; S: sucrose solution; S+OC: sucrose solution supplemented with copaiba oleoresin (200 mg kg^{-1} day^{-1}). Values are the mean ± SD, n = 8. Different superscript letters in a line indicate significant differences between groups according to a Student-Newman-Keuls test. The caloric intake includes energy from sucrose in the drinking water (1.2 kcal mL^{-1}) in the S and S+CO groups.

Variable	Treatments		
	C	S	S+CO
Body weight initial (g)	323 ± 23^{a}	323 ± 24^{a}	323 ± 26^{a}
Body weight final (g)	448 ± 25^{a}	498 ± 24^{b}	444 ± 28^{a}
Body weight gain (g)	125 ± 21^{a}	175 ± 24^{b}	121 ± 37^{a}
Caloric intake (kcal day^{-1})	101 ± 1.9^{a}	108 ± 7.1^{b}	105 ± 0.9^{b}
Epididymal fat (g)	9.84 ± 0.9^{a}	12.8 ± 3.5^{a}	10.1 ± 2.4^{a}
Mesenteric fat (g)	6.18 ± 0.8^{a}	12.9 ± 3.2^{b}	6.84 ± 2.4^{a}
Retroperitoneal fat (g)	14.1 ± 1.8^{a}	19.5 ± 3.5^{b}	13.6 ± 3.8^{a}
Visceral fat (g)	30.1 ± 3.0^{a}	45.3 ± 9.3^{b}	30.7 ± 7.2^{a}
Adiposity index (%)	6.82 ± 0.6^{a}	9.26 ± 1.6^{b}	6.95 ± 1.3^{a}

Table 4. Plasma values of fasting glucose and lipid profile of rats subjected to three treatments for eight weeks. C: control; S: sucrose solution; S+OC: sucrose solution supplemented with copaiba oleoresin (200 mg kg^{-1} day^{-1}). HDL: high density lipoprotein; LDL: low density lipoprotein; VLDL: very low-density lipoprotein; TG: triglycerides. Values are the mean ± SD, n = 8. Different superscript letters in a line indicate significant differences between groups according to a Student-Newman-Keuls test.

Variable	Treatments		
	C	S	S+CO
Fasting glucose (mg dL^{-1})	97.4 ± 12^{a}	112 ± 9^{b}	105 ± 6^{ab}
Total cholesterol (mg dL^{-1})	184 ± 66^{a}	306 ± 117^{b}	227 ± 61^{ab}
HDL (mg dL^{-1})	31.6 ± 11^{a}	32.9 ± 10^{a}	31.1 ± 8^{a}
LDL (mg dL^{-1})	91 ± 41^{a}	172 ± 71^{b}	126 ± 41^{ab}
VLDL (mg dL^{-1})	60.9 ± 22^{a}	102 ± 39^{b}	69.8 ± 23^{a}
Triglycerides (mg dL^{-1})	304 ± 109^{a}	508 ± 194^{b}	349 ± 115^{a}
TG/HDL ratio	10.3 ± 3.2^{a}	17.1 ± 4.3^{b}	12.6 ± 2.1^{a}

Copaiba oleoresin is composed of a high concentration of sesquiterpenes and a small amount of diterpenes (Soares *et al.* 2013). β-caryophyllene, a sesquiterpene with biological activity, is the most studied compound of copaiba oleoresin, and is considered as the source of its anti-inflammatory and antioxidant effects (Fernandez *et al.* 2007; Ames-Sibin *et al.* 2019). The oleoresin used in this study presented *trans*-α-bergamotene as the main compound however, it is important to point out that we cannot exclude the potential contribution of other compounds to our results.

Experimental models of diet-induced obesity are considered appropriate to study obesity and its complications (Nascimento *et al.* 2011; Telles *et al.* 2020; Von Dentz *et al.* 2020). The adopted model, 30% sucrose solution, efficiently induced obesity, as demonstrated by the higher body weight and adiposity index of the rats compared to the control, in agreement with previous studies (Cruz *et al.* 2020; Von

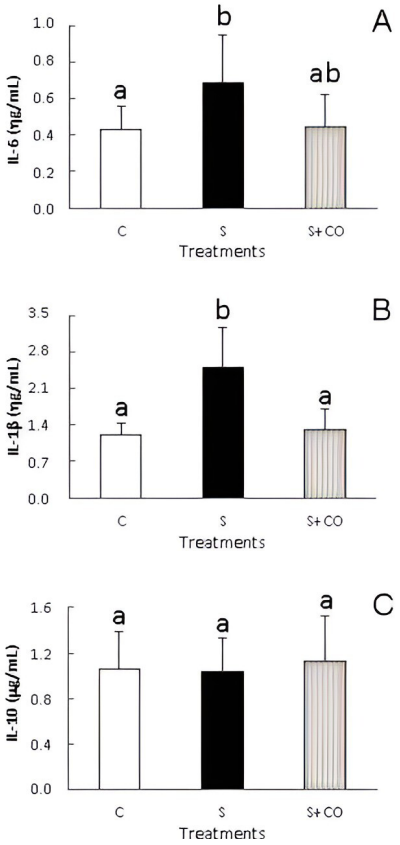

Figure 1. Protein concentration of cytokines in epididymal adipose tissue of rats subjected to three treatments for eight weeks. C: control; S: sucrose solution; S+OC: sucrose solution supplemented with copaiba oleoresin (200 mg kg^{-1} day^{-1}). A – Interleukin (IL)-6; B – IL-1β; C – IL-10. Columns represent the mean and bars the standard deviations, n = 8. Different letters above the bars indicate significant differences between groups according to a Student-Newman-Keuls test. This figure is in color in the electronic version.

Dentz *et al.* 2020). The adiposity index commonly increases because the augmented fat consumption is not accompanied by a proportional increase in fat oxidation, leading to the deposition of fat as TG in the adipose tissue (Rogero and Calder 2018; Longo *et al.* 2019). Adipose tissue responds dynamically to alterations in caloric excess through adipocyte hypertrophy and hyperplasia (Longo *et al.* 2020). Copaiba oleoresin supplementation was efficient in preventing obesity development, inhibiting weight gain and an increase in the adiposity index, in accordance with previous results on the antiobesogenic effect of copaiba (Telles *et al.* 2020). Likewise, the administration of β-caryophyllene (the sesquiterpene present in the copaiba oleoresin) has been shown to decrease the visceral fat index in a high fat/fructose model (Youssef *et al.* 2019). An *in vitro* anti-obesogenic effect of β-caryophyllene showed that it is able to reduce intracellular triglyceride accumulation without interfering with adipocyte number in murine 3T3-L1 adipocytes (Geddo *et al.* 2019).

There is consensus in the literature that obesity is associated with dyslipidemia and hyperglycemia (WHO 2022). In agreement with our results, Cruz *et al.* (2020) showed hypercholesterolemia and hypertriglyceridemia in rats treated with 40% sucrose solution, and suggested sucrose consumption induces hepatic TG synthesis, which could explain the increase in VLDL levels in our study. It is known that adipose tissue dysfunction leads to lipolysis and insulin resistance, resulting in increased hepatic lipogenesis and decreased free fatty acids oxidation, which promotes ectopic liver fat accumulation, leading to the onset of steatosis (Jung and Choi 2014). The TG/HDL ratio is considered a potential biomarker of insulin resistance (Murguía-Romero *et al.* 2013; Wang *et al.* 2017), and its behavior was altered by high sucrose, suggesting insulin resistance in the S group. Copaiba oleoresin supplementation was associated with improvement of the lipid profile and fasting glucose levels. Similarly, 4-week β-caryophyllene administration in an experimental model of high fat/fructose diet was able to ameliorate lipid parameters and fasting blood glucose levels (Youssef *et al.* 2019). The S+CO group presented a lower TG/HDL ratio, indicating prevention of insulin resistance. β-caryophyllene acts on the signaling pathway of insulin release mediated by activation of small G proteins, including Rac1 (Suijun *et al.* 2014). Notably, Rac1 participates in the regulation of type 4 glucose transporter (GLUT4) translocation, favoring the uptake of glucose in insulin-sensitive tissues (Takenaka *et al.* 2016). This could be a possible mechanism by which copaiba oleoresin affects insulin sensitivity. We have to point out that copaiba oleoresin supplementation prevents obesity, which could indirectly induce the improvement in lipid and glucose metabolism and insulin sensitivity.

The obesity condition was also associated with inflammation and altered redox status in the adipose tissue. Among the pro-inflammatory cytokines secreted by dysfunctional adipose tissue evaluated in here, a positive correlation has been reported between IL-6 and adiposity, and it is estimated that white adipose tissue contributes to about one-third of circulating IL-6, with visceral fat producing higher levels (Kwon and Pessin 2013; Rogero and Calder 2018). It is well established that adipose tissue expresses high levels of pro-inflammatory cytokines in obesity, being recognized as an important site of inflammation (Longo *et al.* 2019). Our results are in agreement with other studies showed increased IL-6 in adipose tissue of animals submitted to a high fat and high sugar diet (Francisqueti *et al.* 2017; Von Dentz *et al.* 2020). IL-1β, another important pro-inflammatory cytokine, is induced by the nucleotide-binding domain leucine-rich repeat (NLR) family pyrin domain-containing 3 (NLRP3) inflammasome, and not only impairs peripheral insulin sensitivity, but also interferes with the endocrine and immune functions of adipose tissue (Esser *et al.* 2013; Finucane *et al.* 2015). NLRP3, and consequently IL-1β, is activated in adipose tissue during dietary-induced obesity, and its inhibition significantly alleviates metabolic disorders

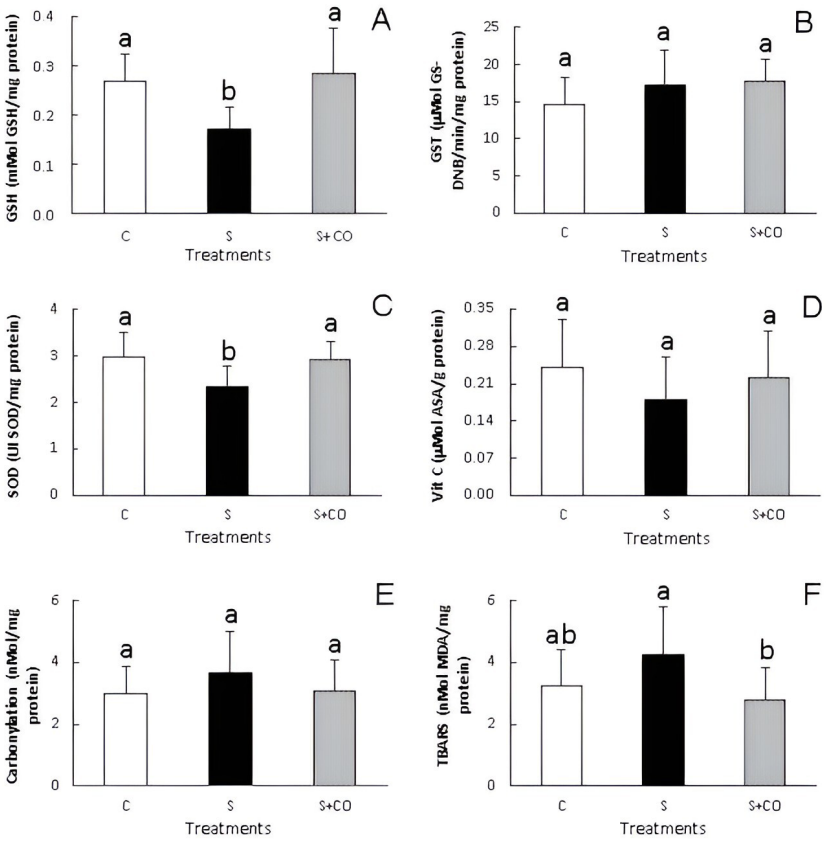

Figure 2. Determination of redox status in epididymal adipose tissue of rats subjected to three treatments for eight weeks. C: control; S: sucrose solution; S+OC: sucrose solution supplemented with copaiba oleoresin (200 mg kg^{-1} day^{-1}). A – reduced glutathione (GSH); B – glutathione S-transferase (GST); C – superoxide dismutase (SOD); D – ascorbic acid (Vit C); E – protein carbonylation; F – thiobarbituric acid reactive substances (TBARS). Columns represent the mean and bars the standard deviations, n = 8. Different letters above the bars indicate significant differences between groups according to a Student-Newman-Keuls test.

(Wu *et al*. 2020). Although copaiba oleoresin did not prevent the increase in IL-6, it was able to minimize it, leading to similar levels as found in the control group. Moreover, copaiba oleoresin supplementation prevented increased IL-1β levels in adipose tissue. β-caryophyllene has anti-inflammatory effects, inhibiting lipopolysaccharide (LPS)-induced TNF and IL-1β expression in peripheral blood (Scandiffio *et al*. 2020). However, to the best of our knowledge, this is the first study to evaluate the effect of copaiba oleoresin on IL-1β levels in adipose tissue. Despite the observed beneficial effects on inflammation, the mechanism by which copaiba oleoresin acts on adipose tissue remains unknown.

Dietary patterns that drive obesity play a central role in inflammation and oxidative stress in adipose tissue, as hypertrophic adipocytes induce the synthesis of pro-inflammatory cytokines, which promote increased generation of reactive oxygen and nitrogen species by macrophages and monocytes (Engin 2017). On the other hand, reactive oxygen species (ROS) induce the further release of pro-inflammatory cytokines through redox-sensitive transcription factors, particularly NF-κB and the NADPH oxidase pathway (Marseglia *et al*. 2015; Engin, 2017). Also, ROS are essential for activation of the inflammasome (Zielinska-Blizniewska *et al*. 2019). Thus, there is a clear feedback interplay between inflammation and oxidative stress in obesity.

Oxidative damage is greater in obese subjects, in part, because of depleted antioxidant sources (Marseglia *et al*. 2015). Correspondingly, in this study, the high sucrose diet decreased SOD and GSH activity in adipose tissue. SOD and GSH are antioxidant enzymes which *play an* indispensable role in the antioxidant protective capacity of biological systems against free radical attack (Ighodaro and Akinlove 2018). Copaiba oleoresin supplementation led to antioxidant effects in adipose tissue, preventing the decrease in SOD and GSH activity, and reducing TBARS levels compared to the high sucrose fed rats. Corroborating the beneficial effects of copaiba oleoresin on redox status, Telles *et al*. (2020) showed copaiba oleoresin supplementation was associated to higher levels of GST, SOD and Vit C, and decreased hepatic carbonylated protein levels in a high sucrose model. Copaiba oleoresin reduced carbonylated protein and ROS, and increased GSH levels from arthritic rats, improving the oxidative status by decreasing the inflammatory process, stimulating the endogenous antioxidant system, or acting as a direct free radical scavenger (Ames-Sibin *et al*. 2019).

CONCLUSIONS

Copaiba oleoresin induced an antiobesogenic effect in Wistar rats, which was associated to improvement in inflammation and redox status in adipose tissue of animals summitted to high sucrose diet. Although the adopted diet model did mimic the clinical condition, it gives no information regarding as to whether these findings are applicable to humans or not. However, it addresses important benefits by using non-pharmacological therapy based on a natural product. This study provides new information regarding the effects of copaiba oleoresin on the adipose tissue, suggesting its use could be a promising approach against obesity and its complications, such as adipose tissue inflammation and redox imbalance.

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