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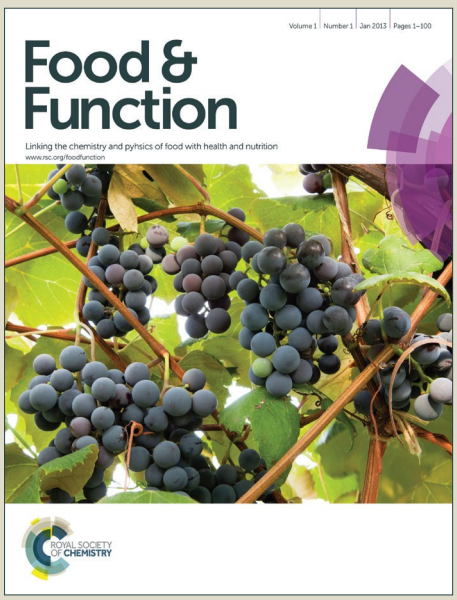

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Cinnamon extract improves the body composition and attenuates lipogenic processes in the liver and adipose tissue of rats

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Abstract

In models of metabolic disorders, cinnamon improves glucose and lipid metabolism. This study explores the effect of chronic supplementation with aqueous cinnamon extract (CE) on the lipid metabolism of rats. Male adult Wistar rats were separated into a control group (CTR) receiving water and a CE Group receiving aqueous cinnamon extract (400 mg of cinnamon/kg body mass/day) by gavage for 25 consecutive days. Cinnamon supplementation did not change the food intake or the serum lipid profile but promoted the following changes: lower body mass gain (P=0.008), lower relative mass of white adipose tissue (WAT) compartments (P=0.045) and higher protein content (percentage of the carcass) (P=0.049). The CE group showed lower leptin mRNA expression in the WAT (P=0.0017) and an important tendency for reduced serum leptin levels (P=0.059). Cinnamon supplementation induced lower mRNA expression of SREBP1c (sterol regulatory element-binding protein 1c) in the WAT (P=0.001) and liver (P=0.013) and lower mRNA expression of SREBP2 (P=0.002), HMGCoA reductase (3-hydroxy-3-methylglutaryl-CoA reductase) (P=0.0003), ACAT1 (acetyl-CoA acetyltransferase 1) (P=0.032) and DGAT2 (diacylglycerol O-acyltransferase 2) (P=0.03) in the liver. These changes could be associated with the reduced esterified cholesterol and triacylglycerol content detected in this tissue. Our results suggest that chronic ingestion of aqueous cinnamon extract attenuates lipogenic processes, regulating the expression of key enzymes and transcriptional factors and their target genes, which are directly involved in lipogenesis. These molecular changes possibly promote adaptations that would prevent an increase in circulating cholesterol and triacylglycerol levels and prevent lipid accumulation in tissues, such as liver and WAT. Therefore, we speculate that cinnamon may also be useful for preventing or retarding the development of lipid disorders.

Keywords *Cinnamomum zeylanicum*; lipid metabolism; triacylglycerol; cholesterol; liver; white adipose tissue.

Abbreviations

LDL; low-density lipoprotein

WAT; white adipose tissue

SREBPs; Sterol Regulatory Element-Binding Proteins

PPARs; Peroxisome Proliferator-Activated Receptors

CE; cinnamon water extract

HDL; high-density lipoprotein

LDLR; Low-Density Lipoprotein Receptor

HMGCoA reductase; 3-hydroxy-3-methylglutaryl-CoA reductase

CYP7A1; Cholesterol 7 alpha-hydroxylase

ACAT1; acetyl-CoA acetyltransferase 1

DGAT2; diacylglycerol O-acyltransferase 2

TAG; triacylglycerol

TLC; Thin layer chromatography

Introduction

Cinnamon (*Cinnamomum zeylanicum*), an ancient medicinal herb traditionally harvested in Asians countries, is composed of a variety of bioactive compounds, such as polyphenols, cinnamaldehyde and flavonoids; these compounds exhibit beneficial effects, including potent antioxidant, anti-inflammatory and anti-diabetic properties.[1] Studies in humans and experimental models of obesity and type 2 diabetes mellitus have shown that cinnamon consumption was associated with improved glucose metabolism,[2] reduced body fat[2,3] and reduced serum levels of triglycerides, total cholesterol and low-density lipoprotein (LDL).[4,5]

Because of its positive effects on metabolism, cinnamon supplementation has been proposed for the treatment of lipid or glucose disorders. The vast majority of these studies were performed in patients with or animal models of metabolic diseases and little is known about the effect of cinnamon consumption on individuals with no established disease. There are very few studies in healthy humans suggesting that cinnamon has beneficial effects on glucose and lipid metabolism.[6,7] Therefore, it is uncertain whether cinnamon can be useful in preventing or retarding the development of metabolic disturbances.

Moreover, although some potential mechanisms for the action of cinnamon have been identified,[5,8,9,10] the molecular characterization of the mechanisms by which cinnamon exerts its metabolic effects, especially regarding the lipid metabolism, remains to be fully clarified. In the present study, we analyzed the effects of chronic supplementation with an aqueous cinnamon extract on body mass and composition, key steps of lipid metabolism pathways in the liver and adipose tissue of rats and the hormonal and molecular factors underlying the actions of cinnamon.

Materials and Methods

Animals and experimental design

The Institutional Animal Care Committee of Fluminense Federal University approved the experimental design (107/2009). Three-month-old adult male Wistar rats were maintained under controlled temperature conditions (24±1°C) on a 12 h cycle of alternating darkness and artificial light (lights on at 7:00 am). The standard chow (Nuvilab, Rio de Janeiro, Brazil) and water were provided *ad libitum*. The macronutrient content of the standard chow is 22% of protein, 4,5% of fat and 60% of carbohydrate, approximately (% per kg).

The animals were randomly separated into two groups, Control Group (CTR; n=6) receiving water by gavage and Aqueous Cinnamon Extract Group (CE; n=4) receiving aqueous cinnamon extract by gavage; both treatments were administered for 25 days.

All the experiments were performed using the same batch of cinnamon bark (*Cinnamomum zeylanicum),* obtained from a local producer. The protocol of extract preparation was adapted from Kannappan and colleagues[11] and Sheng and colleagues.[5] The extract obtained was analyzed by high performance liquid chromatography (DAD-HPLC-UV) and (+)-ESI-MS and (-)-ESI-MS. The HPLC analysis showed that the major peak obtained is characteristic of the chemical marker cinnamaldehyde, a very common phytochemical in the *Cinnamomum genus* (data not shown).[12] In addition to cinnamaldehyde, other substances were identified by ESI-MS such as phenylpropanoids, fatty acids and procyanidins.

The rats received 4 mL of aqueous cinnamon extract/kg body mass/day (corresponding to 400 mg of cinnamon/kg body mass/day). This dose was based on the study of Sheng *et al*.[5] The adult human equivalent dose is 64 mg/kg per day, based on the body

surface area normalization method for the translating the drug doses used in animal studies to appropriate doses for human consumption.[13]

Body mass and chow intake were measured three and two times a week, respectively. Rats in the fed state were sacrificed by decapitation, and both the subcutaneous (inguinal) and visceral (retroperitoneal and epididymal) WAT compartments were excised and weighed. Tissue samples (80 mg) were harvested and stored at -70°C. The remaining adipose tissue was returned to the carcasses, which were eviscerated, weighed and frozen at -20°C for body composition analysis. Blood obtained from the trunk was centrifuged (1700xg for 15 min at 4°C), and the resulting serum was stored at -20°C until analysis.

Basal glycemia

Seventy-two hours before being sacrificed, the rats were fasted for 12 h for a blood glucose measurement using a glucometer (ACCU-Check Advantage, Roche), which was based on the glucose-glucose oxidase reaction. The results are expressed in mg/dL.[14]

Body composition analysis

The fat and protein contents of the carcasses were determined as previously described.[15] Briefly, eviscerated carcasses were weighed, autoclaved and homogenized in distilled water (1:1). Aliquots of homogenate were used for the measurement of protein and fat contents. Aliquots of homogenate were used to determine fat mass gravimetrically. Samples were hydrolyzed in a shaking water bath at 70°C for 2 h with 30% KOH and ethanol. After the addition of 9 M sulfuric acid, total lipids were extracted by three successive washes with petroleum ether (Vetec, Rio de Janeiro, RJ, Brazil). The samples were dried at room temperature until constant weight had been obtained. Protein was extracted from homogenates using 0.6 N KOH at 37°C for 1 h. After that, the samples were centrifuged at 800×g for 10

min, and the supernatant was collected to measure protein concentration by employing Bradford reagent. The results are expressed as grams of fat or protein per 100 g of carcass.

Hormone measurements

The serum adiponectin, leptin and insulin concentrations were measured using specific rodent radioimmunoassay kits (Linco Research, MO, USA) in accordance with the manufacturer´s instructions. The sensitivity was 0.78 μg/mL for adiponectin, 0.5 ng/mL for leptin and 0.1 ng/mL for insulin. All samples were measured using the same assay, and the intra-assay variation was 4.5% for adiponectin, 8.2% for leptin, and 6.4% for insulin.

Serum lipid profile

Measurements of the total cholesterol, high-density lipoprotein (HDL) cholesterol and triglycerides (TG) were performed using commercial kits from Biosystems (Paraná, Brazil) and an automated A15 spectrophotometer (Biosystems S.A., Barcelona, Spain). The serum very-low-density lipoprotein (VLDL) cholesterol and LDL cholesterol were estimated according to Friedwald Equation: VLDL - cholesterol = TG/5; LDL - cholesterol = [total cholesterol - HDL-cholesterol –TG]/5.[16]

Thin layer chromatography (TLC)

Lipid extraction was performed using the method of Bligh and Dyer[17] with modifications. Serum (5000 μg of protein), liver and white adipose tissue samples (10 mg) were mixed for 2 hours in a chloroform-methanol-water solution (2:1:0.8 v/v) with intermittent shaking. The mixture was centrifuged (1500 x g for 20 minutes at 4°C) in a Sorvall RC-5b preparative centrifuge (Sorvall Centrifuge, Newtown, CT, USA), the supernatant was collected, and chloroform was added. The mixture was shaken, and after

centrifugation (1500 x g for 20 minutes), the organic phase was removed and dried under nitrogen. The extracted lipids were analyzed by one-dimensional TLC for neutral lipids. To visualize the lipids, plates were immersed in a charring solution consisting of 3% $CuSO_4$ and 8% H_3PO_4 (v/v) for 10 s and heated to 110°C for 10 minutes.[18] The charred TLC plates were then analyzed by densitometry (ImageMaster TotalLab software, USA).

mRNA expression analysis

Total liver RNA was isolated from samples using Trizol reagent (Invitrogen, CA, USA); a commercial RNeasy lipid tissue mini kit (Qiagen, TX, USA) was used for epididymal adipose tissue RNA. The total RNA was reverse transcribed using 1 μg of RNA and a Superscript III kit (Invitrogen, CA, USA). The mRNA expression was evaluated by real-time PCR using specific primers, and 36B4 was used as a reference gene (Table 1). The products were amplified on an Applied Biosystems 7500 Real-Time PCR System (Life Technologies Corp., CA, USA) using SYBR Green PCR Master Mix (Applied Biosystems, CA, USA). The cycle parameters were 50°C for 2 min and 95°C for 10 min, followed by 40 cycles of 95°C for 15 s, 60°C for 30 s, and 72°C for 45 s. The relative mRNA expression ($2^{\Delta\Delta Ct}$) was calculated by comparing the cycle threshold (Ct) between groups after correction for the reference gene 36B4. The expression of the 36B4 gene was stable for both tissues and experimental groups. The efficiency of each reaction was calculated using a serial dilution and varied from 95% to 105%. Each sample was measured in duplicate, and the results are expressed relative to the values of the control group, which was set to 1. The purity of the PCR products was checked by analyzing the melting curves.

Statistical analyses

Data are expressed as the means ± standard error of the mean (S.E.M.) and were analyzed by Student's t test using GraphPad Prism 5 (GraphPad Prism Software, Inc., CA, USA). Differences were considered significant at $P<0.05$.

Results

As shown in Figure 1A, the oral administration of cinnamon extract to healthy rats led to a lower body mass gain ($P=0.008$) with no significant changes in the cumulative food intake compared with that of the control (Figure 1B). The total WAT mass (subcutaneous and visceral compartments) adjusted for the body mass of the CE group was lower than that of CTR (Figure 1C, $P=0.045$). The body composition analysis revealed a higher protein content in the carcasses of the CE rats (Figure 1D, $P=0.049$); importantly, the lipid content of their carcasses was also lower (41.4%, Figure 1E, $P=0.09$).

The lower total WAT mass was accompanied by a lower serum leptin level and a substantial decrease in the leptin mRNA expression in the WAT of the CE group (Figure 2A, $P=0.059$ and Figure 2B, $P=0.0017$). Compared with the control, the CE-treated rats had lower serum insulin levels (Figure 2C, $P=0.007$) without changes in the basal glycemia (Figure 2D) and the serum adiponectin levels (Figure 2E).

Although cinnamon extract did not change the serum lipid profile (Table 2), in the liver the treatment induced a lower esterified cholesterol content (Figure 3A, $P=0.010$) along with a lower expression of the mRNA for acetyl-CoA acetyltransferase 1 (ACAT) (Figure 3B, $P=0.032$). In addition, the CE-treated rats exhibited a lower mRNA expression of the transcriptional regulator SREBP2 and its target gene, 3-hydroxy-3-methylglutaryl-CoA reductase (HMGCoA reductase) (Figure 3C, $P=0.002$ and Figure 3D, $P=0.0003$). However, no significant changes in the mRNA expression of PPARα and its target genes, CYP7A1 and LDLR, (Figure 3E, F and G) were observed in the CE-treated rats.

As depicted in figure 4A, the cinnamon extract-treated rats showed a lower hepatic triacylglycerol content (P=0.0208) without changes in the monoacylglycerol and phospholipid contents (data not shown). In addition, the mRNA expression of diacylglycerol O-acyltransferase 2 (DGAT2) and SREBP1c were lower in the CE-treated rats (Figure 4B, P=0.03 and Figure 4C, P=0.013).

In the WAT, the CE group exhibited a lower content of triacylglycerol (Figure 5A, P=0.0058) and lower mRNA expression of SREBP1c (Figure 5B, P=0.0010), without changes in the monoacylglycerol and phospholipid contents (data not shown).

Discussion

In the present study, we demonstrated that chronic supplementation with aqueous cinnamon extract improves the body composition, reducing the body mass and the fat depot mass without changes in the cumulative food intake of rats. These data corroborate recent findings.[19,20] that showed reduced body mass gain with no food intake changes in healthy or high-fat-sucrose diet-fed rats treated with cinnamon extract for 30 or 56 days. However, other studies using different doses and time courses of cinnamon treatment in rodent models reported no changes in body mass gain.[3,5, 21]

The lower body mass gain in the CE-treated rats was accompanied by a lower relative WAT mass. Reduced fat mass depots were previously found in obese rodents treated with cinnamon.[3,9,19] In addition, the decreased fat mass and increased protein content in the carcasses of the CE group indicate that chronic treatment with cinnamon extract improves the body composition. Ziegenfuss and colleagues[2] reported the same beneficial effects on body composition in pre-diabetic humans treated with cinnamon. However, to the best of our knowledge, there are no data regarding the effects of cinnamon on the lean body mass of

experimental animals. Therefore, our results indicate that treatment with an aqueous cinnamon extract results in a better body composition in healthy animals.

The most frequently reported beneficial effect of supplementation with cinnamon is improved insulin sensitivity.[3,4] In our study, we observed lower serum insulin levels and normal basal glycemia in the CE-treated rats, suggesting higher insulin sensitivity. Insulin sensitivity is strongly related to the serum levels of adiponectin[8], an adipocyte-derived hormone; however, in the present study, the aqueous cinnamon extract had no effect on the serum adiponectin levels. The adiponectin response to cinnamon treatment has been debated, and previous reports showed either no effect in fructose-fed rats [10] or an increase in the serum adiponectin level in *db/db* mice;[9] in contrast, a strong inhibition has been reported in the 3T3-L1 cell line. Therefore, altogether, the data suggest that the effect of cinnamon on insulin sensitivity is, at least in part, independent of an adiponectin increase.

Leptin, another important adipocytokine, is a major regulator of food intake and energy balance.[22] We observed a marked reduction in the serum leptin concentrations and the leptin mRNA expression in the adipose tissue of cinnamon-treated rats. These data can be explained, at least in part, by the lower WAT mass observed in these animals because the serum leptin level is positively correlated with adipose tissue mass;[23] however, these data may also be related to a direct effect of cinnamon on leptin gene expression. This latter hypothesis is supported by the study of Cao and colleagues,[8] who showed that cinnamon suppressed the leptin gene expression in 3T3-L1 adipocytes. Interestingly, although leptin is an anorexigenic hormone, the cinnamon-treated animals did not show any changes in food consumption.

Cinnamon reduces the total cholesterol, LDL cholesterol and triglyceride levels in patients with type 2 diabetes[4] or nonalcoholic fatty liver disease[24] and has hypolipidemic effects in obese rodents.[5,11] However, our study shows that supplementation with cinnamon

did not affect the serum lipid profile of healthy animals, suggesting that the hypolipidemic effect of cinnamon can be more easily observed in conditions of dyslipidemia.

Regardless of the lack of alterations in the serum lipid levels, the cinnamon treatment produced important changes in the hepatic metabolism of cholesterol and triglycerides. For the first time, we showed that cinnamon extract treatment induced a reduction in the liver content of esterified cholesterol that was accompanied by lower expression of ACAT, a key enzyme in the esterification of cholesterol.[25] This finding is interesting because the inhibition of ACAT is a well-known mechanism for reducing plasmatic circulating cholesterol levels since esterification is essential to cholesterol transport in lipoproteins. Indeed, some studies suggest that the inhibition of ACAT is an attractive method for treating hyperlipidemia.[25] Furthermore, the lower mRNA expression of SREBP2 and its target gene HMGCoA reductase, which is the rate-limiting enzyme in cholesterol biosynthesis,[26] may contribute to the phenotype observed in the liver of the CE-treated animals. However, we did not observe any changes in the hepatic PPARα mRNA expression or the expression of its target genes LDLR and CYP7A1; these targets encode the LDL receptor and cholesterol 7α hydroxylase, respectively, which are key proteins in cholesterol metabolism.[27,28,29] Our data regarding PPARα expression differs from that of other studies in animal models exhibiting dyslipidemia,[24] which showed increased PPARα gene expression associated with improvements in the serum lipid profile of cinnamon-treated animals. To the best of our knowledge, there are no studies describing the effects of cinnamon on LDLR and CYP7A1 gene expression. Therefore, the lack of up-regulation of these genes in our study could contribute to the unaltered levels of serum cholesterol in healthy rats.

Our findings clearly indicate that cinnamon supplementation induces a propitious adaptation in hepatic and adipose tissue lipid metabolism that favors a lower accumulation of triglycerides. These changes were accompanied by a relevant reduction in the mRNA

expression of SREBP1c, which is a master transcriptional regulator of fatty acid and de novo TAG synthesis[30] and is highly responsive to insulin[31] in WAT and liver. These data corroborate a previous study in a fructose-fed rat model, which showed that cinnamon extract supplementation reduced the SREBP1c expression in WAT.[10]

In the liver of the CE animals, we identified a new target for cinnamon. The lower content of TAG was associated with a lower expression of DGAT2, the enzyme that catalyzes the final step in triglyceride synthesis.[32] Studies suggest that the suppression of DGAT2 associated with lower SREBP1c expression has a protective effect against insulin resistance and liver steatosis.[32] This process may be a novel mechanism by which cinnamon supplementation can improve insulin sensitivity.

Conclusions

In conclusion, our results suggest that chronic supplementation with aqueous cinnamon extract attenuates lipogenic processes, regulating the expression of transcriptional factors and their target genes that are directly involved in lipogenesis. These molecular changes possibly promote adaptations that could prevent the increase in circulating cholesterol levels and the lipid accumulation in tissues, such as liver and adipose tissue. Therefore, our study supports the suggestion that cinnamon is an efficient nutraceutical for the treatment of lipid disorders, and we speculate that cinnamon may also be useful for preventing or retarding the development of lipid disorders.

Competing interests

The authors declare that there is no conflict of interest.

Authors' contributions

The authors' contributions were as follows: Bruna P. Lopes, Carmen C. Pazos-Moura and Karen J. Oliveira designed the research project and wrote the manuscript. Bruna P. Lopes, Thaiane G. Gaique, Luana L. Souza, Gabriela S. M. Paula, Tania M. Ortiga- Carvalho and Karen J. Oliveira performed the experimental and most of the laboratory procedures, and performed the statistical analyses. Anne Caroline C. Gomes, Naomi K. Simas, Ricardo M. Kuster performed the cinnamon extract analysis. Bruna P. Lopes, George E. G. Kluck and Georgia C. Atella performed the TLC analysis. All authors read and approved the final content.

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Figures

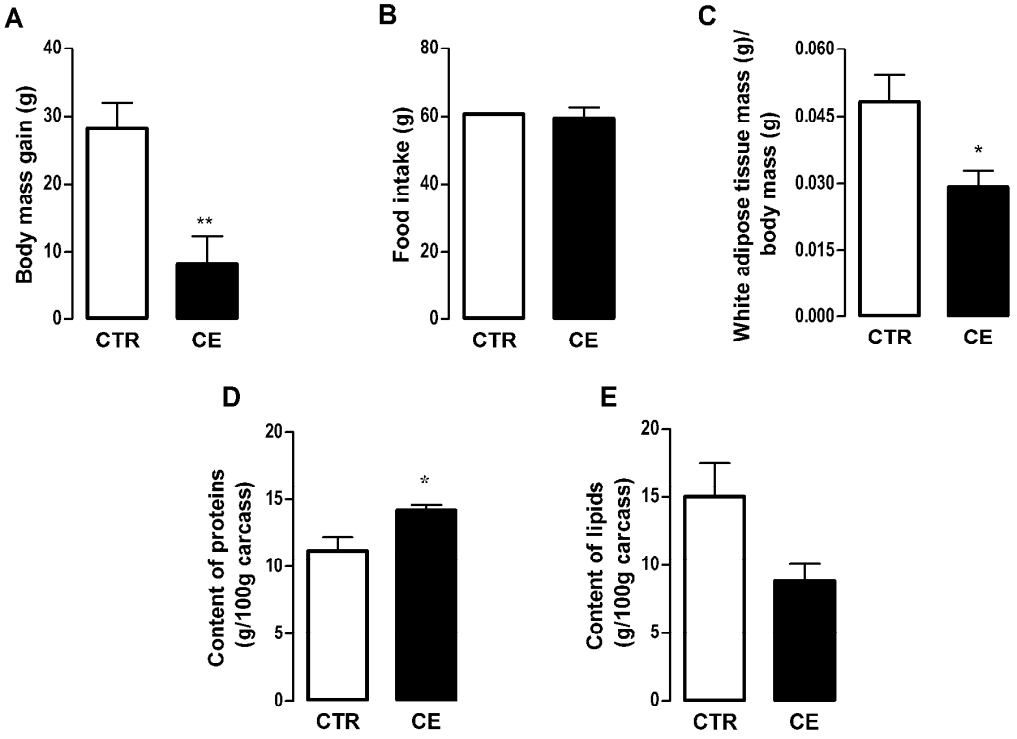


Fig. 1 Body mass gain (A), cumulative food intake (B), white adipose tissue mass (C), protein content (percentage of the carcass) (D) and lipid content (percentage of the carcass) (E) of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR). CTR: n=6; CE: n=4. *$P<$ 0.05; **$P<$ 0.01. Values are expressed as the means ± S.E.M.

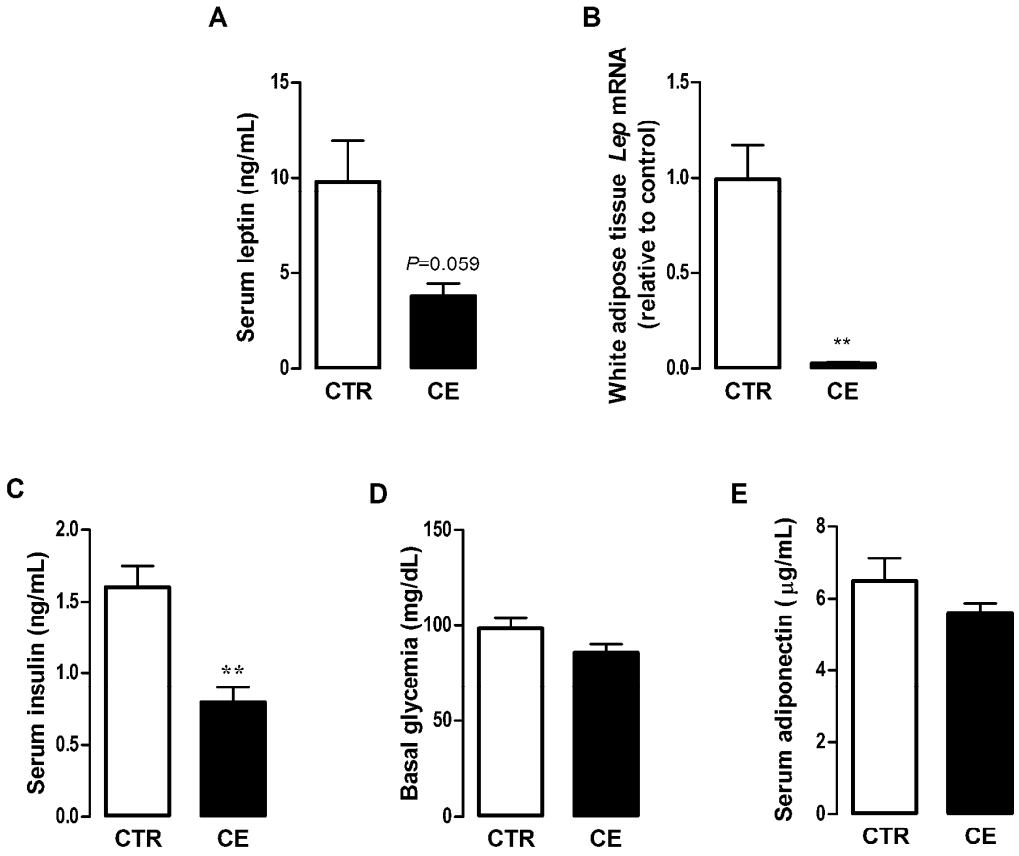


Fig. 2 Serum leptin (A), white adipose tissue leptin (*Lep*) mRNA expression (B), serum insulin (C), Basal glycemia (D) and serum adiponectin (E) of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR). *Lep* mRNA results were normalized by the reference gene 36B4 and expressed relative to those of the control group. CTR: n=6; CE: n=4. $^{**}P < 0.01$. Values are expressed as the means ± S.E.M.

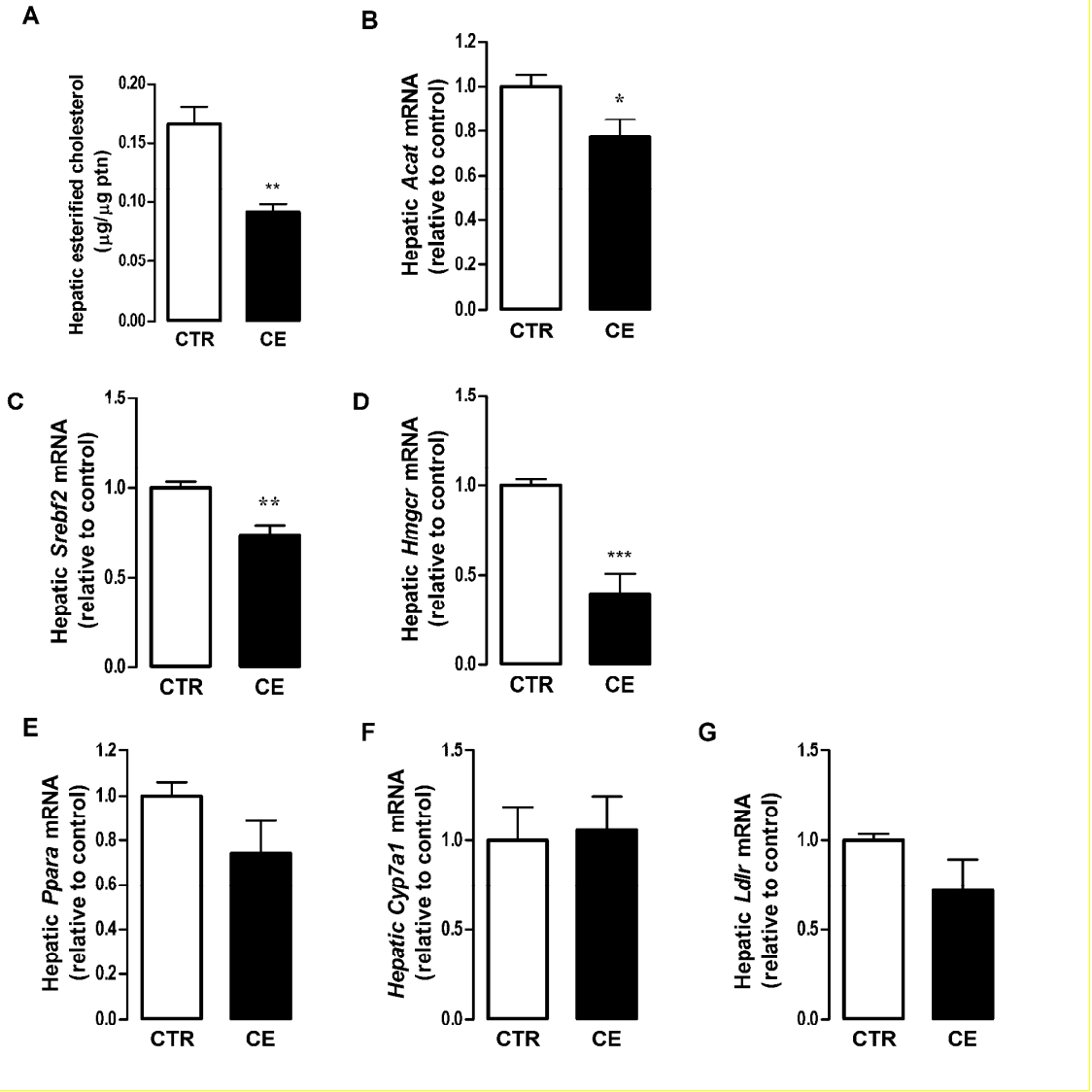


Fig. 3 Cholesterol metabolism in liver of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR). Esterified cholesterol content (A), mRNA expression of *Acat* (acetyl-CoA acetyltransferase 1, ACAT1) (B), *Srebpf2* (sterol regulatory element-binding protein 2, SREBP2) (C), *Hmgcr* (3-hydroxy-3-methylglutaryl-CoA reductase, HMGCoA reductase) (D), *Ppara* (peroxisome proliferator-activated receptor, PPARα) (E), *Cyp7a1* (cholesterol 7 alpha-hydroxylase, CYP7A1) (F) and *Ldlr* (low-density lipoprotein receptor, LDLR) (G). mRNA results were normalized by the reference gene 36B4 and expressed relative to those of the

control group. CTR: n=6; CE: n=4. *P<0.05; **P< 0.01. Values are expressed as the means ± S.E.M.

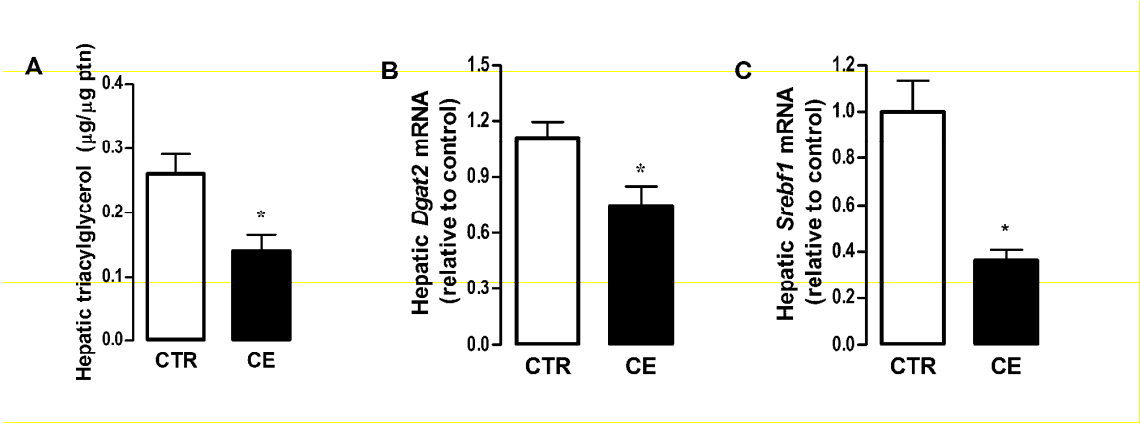


Fig. 4 Triacylglycerol metabolism in liver of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR). Triacylglycerol content (A), mRNA expression of *Dgat2* (diacylglycerol O-acyltransferase 2, DGAT2) (B) and *Srebf1* (sterol regulatory element-binding protein 1c) (C). mRNA results were normalized by the reference gene 36B4 and expressed relative to those of the control group. CTR: n=6; CE: n=4. *P<0.05; **P< 0.01. Values are expressed as the means ± S.E.M.

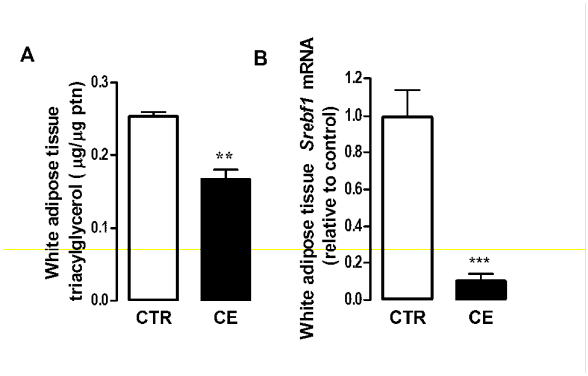


Fig. 5 Triacylglycerol metabolism in white adipose tissue of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR). Triacylglycerol content (A) and *Srebf1* (Sterol Regulatory Element-Binding Protein 1c) (B) mRNA expressed relative to the control group, after normalization by the reference gene 36B4. CTR: n=3; CE: n=3. ***P*< 0.01, ****P*< 0.001. Values are expressed as the means ± S.E.M.

Table 1 Primer sequences

Protein	Gene	Seqnence	Reference number
Leptin	*Lep*	Forward 5'-CATCTGCTGGCCTTCTCCAA-3' Reverse 5'-ATCCAGGCTCTCTGGCTTCTG-3'	14
PPARα	*Ppara*	Forward 5'-TTCAATGCCCTCGAACTGGA-3' Reverse 5'-GCACAATCCCCTCCTGCAAC-3'	33
SREBP2	*Srebf2*	Forward 5'- CACTCACGCTCCTCGGTCAC-3' Reverse 5'-CGGATAAGCAGGTCTGTAGGTTGG-3'	34
SREBP1c	*Srebf1c*	Forward 5'-AAAACCAGCCTCCCCAGAGC-3' Reverse 5'-CCAGTCCCCATCCACGAAGA-3'	33
LDLR	*Ldlr*	Forward 5'-CCAGTGCGGCGTAGGATT-3' Reverse 5'-GGGACTCATCGGAGCCAT-3'	35
CYP7A1	*Cyp7a1*	Forward 5'-TGCTCTGTCTTCACTTTCTG-3' Reverse 5'-ACTCGGTAACAGAAGGCATA-3'	36
ACAT 1	*Acat*	Forward 5'-GCTGAAGTGAACTACCCCTT -3' Reverse 5'-GAGCCATGCCTCTAGTACCT-3'	37
DGAT2	*Dgat2*	Forward 5'-AGGCCTTGATGGTTTCTATCCA -3' Reverse 5'- GCTGCCCTTCCCCAATTAAC-3'	38
HMGCoA reductase	*Hmgcr*	Forward 5'CCAGGATGCAGCACAGAATGT -3' Reverse 5'- CCAATTCGGGCAAGCTGCCG -3'	26
36B4	*Rplp0*	Forward 5'- TTCCCACTGGCTGAAAAGGT-3' Reverse 5'-CGCAGCCGCAAATGC-3'	15

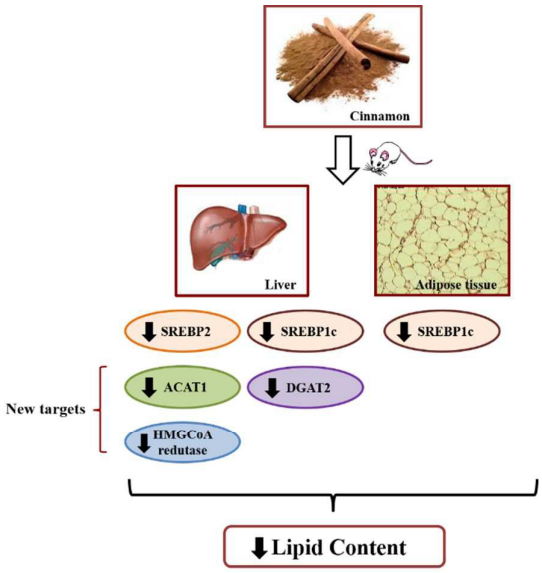
PPAR: Peroxisome proliferator-activated receptor; SREBP: Sterol regulatory element binding protein; LDLR: Low-density lipoprotein receptor; CYP7A1: Cholesterol 7α hydroxylase; ACAT: Acetyl CoA acetyltransferase 1; DGAT2: Diacylglycerol O-acyltransferase 2; and HMGCoA reductase: 3-hydroxy-3-methylglutaryl-CoA reductase.

Table 2 Serum lipid profile of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR).

	CTR		CE	
	Mean	SEM	Mean	SEM
Total cholesterol (mg/dL)	97.25	15.46	83.50	4.77
HDL (mg/dL)	17.50	2.47	15.75	1.10
LDL (mg/dL)	57.50	10.65	45.75	7.36
V_DL (mg/dL)	20.00	1.97	22.00	4.02
Triglycerides (mg/dL)	94.33	9.65	110.3	19.36
Free fatty acids (ng/μg protein)	15.00	0.6831	15.75	1.315

Data were obtained using commercial kits (triglycerides, total cholesterol and fractions) and thin layer chromatography (free fatty acids). HDL: high density lipoprotein; LDL: low density lipoprotein; VLDL: very low density lipoprotein; CTR: *n=4-6*; CE: *n=4*. Values are expressed as the means ± S.E.M.

Cinnamon attenuates lipogenic processes, regulating the expression of key enzymes, transcriptional factors and their target genes in liver and adipose tissue.



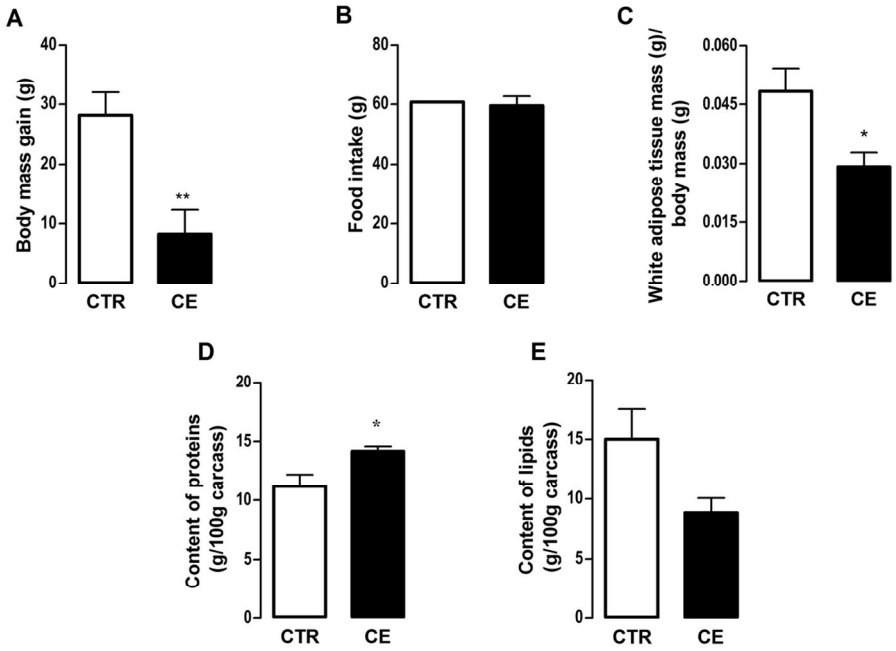


Fig. 1 Body mass gain (A), cumulative food intake (B), white adipose tissue mass (C), protein content (percentage of the carcass) (D) and lipid content (percentage of the carcass) (E) of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR). CTR: n=6; CE: n=4. *P< 0.05; **P< 0.01. Values are expressed as the means ± S.E.M.
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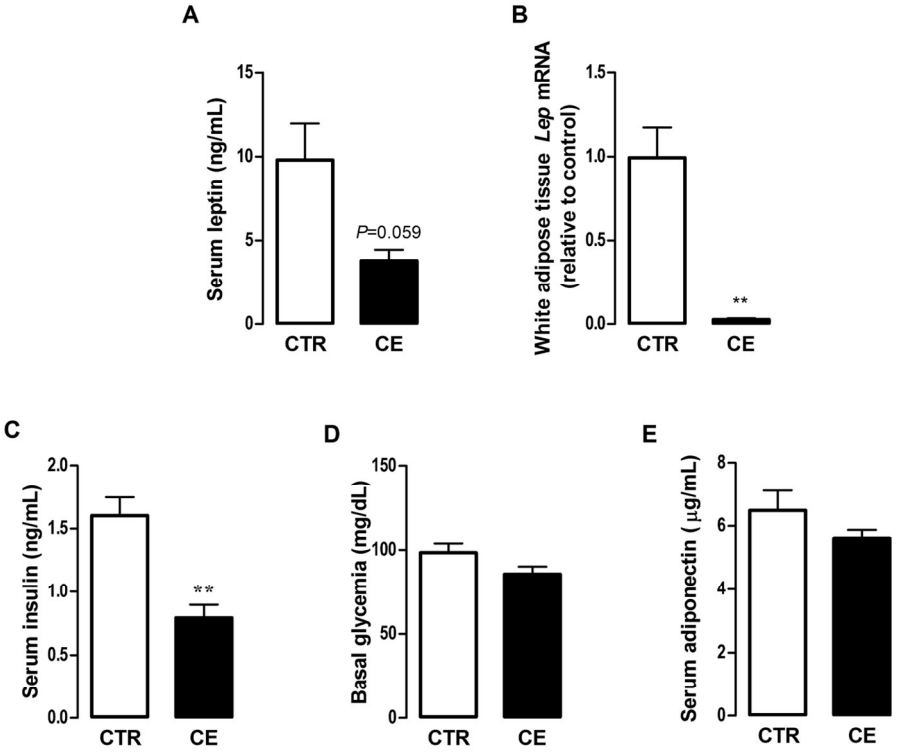

Fig. 2 Serum leptin (A), white adipose tissue leptin (Lep) mRNA expression (B), serum insulin (C), Basal glycemia (D) and serum adiponectin (E) of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR). Lep mRNA results were normalized by the reference gene 36B4 and expressed relative to those of the control group. CTR: n=6; CE: n=4. **P< 0.01. Values are expressed as the means ± S.E.M.
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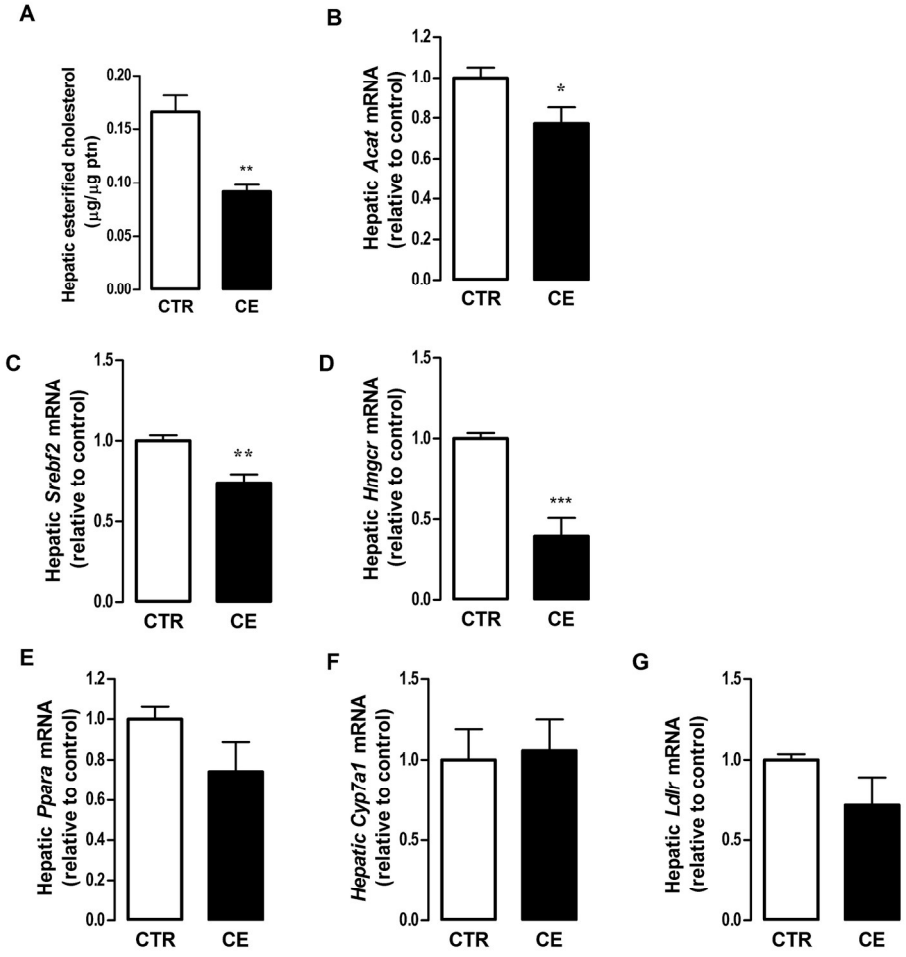
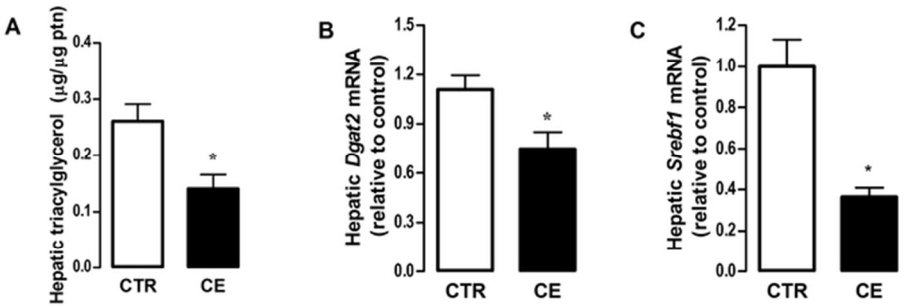


Fig. 3 Cholesterol metabolism in liver of rats treated with Aqueous Cinnamon Extract (CE) and Controls (CTR). Esterified cholesterol content (A), mRNA expression of Acat (acetyl-CoA acetyltransferase 1, ACAT1) (B), Srebpf2 (sterol regulatory element-binding protein 2, SREBP2) (C), Hmgcr (3-hydroxy-3-methylglutaryl-CoA reductase, HMGCoA reductase) (D), Ppara (peroxisome proliferator-activated receptor, PPARα) (E), Cyp7a1 (cholesterol 7 alpha-hydroxylase, CYP7A1) (F) and Ldlr (low-density lipoprotein receptor, LDLR) (G). mRNA results were normalized by the reference gene 36B4 and expressed relative to those of the control group. CTR: n=6; CE: n=4. *P<0.05; **P< 0.01. Values are expressed as the means ± S.E.M.
171x171mm (300 x 300 DPI)



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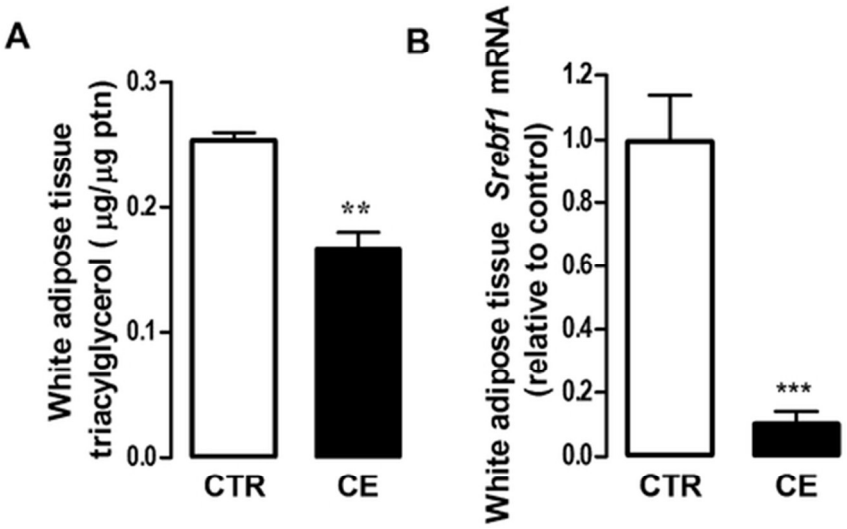

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52x33mm (300 x 300 DPI)

Table 1 Primer sequences

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