BASIC AND EXPERIMENTAL NUTRITION
DOI: 10.12957/demetra.2025.84467

DEMETRA
Alimentação, Nutrição & Saúde

e-ISSN:2238-913X

Eveline Costeira Bálsamo[1]
Luiggi Muller Madalosso[1]
Bianca dos Santos Bertolazi[1]
Hecson Jesser Segat[1]
Silvana Peterini Boeira[1]

[1] Universidade Federal do Pampa, Curso de Nutrição, Laboratório de Avaliações Farmacológicas e Toxicológicas aplicadas à Moléculas Bioativas do Pampa. Itaqui, RS, Brasil.

Manuscript from the Final Course Project entitled "Hypolipidemic effect of cinnamon extract (Cinnamomum zeylanicum) in a model of hypercholesterolemia," authored by Eveline Costeira Bálsamo and supervised by Silvana Peterini Boeira, defended as a requirement for obtaining the title of Nutritionist from the Universidade Federal do Pampa (Federal University of Pampa) – Itaqui Campus. December 2017.

Correspondence
Silvana Peterini Boeira
silvanaboeira@unipampa.edu.br

Assistant Editor:
Luciane Pires da Costa

Hypolipidemic effect of cinnamon extract (Cinnamomum zeylanicum) in a hypercholesterolemia model

Efeito hipolipemiante do extrato de canela (*Cinnamomum zeylanicum)* em um modelo de hipercolesterolemia

Abstract

Introduction: Hypercholesterolemia is the main risk factor for the formation of atheromatous plaques in the arteries and is an important risk factor for the development of Alzheimer's disease. Among the plants that have been the subject of various pharmacological studies, *cinnamon* (*Cinnamomum zeylanicum*) stands out as having natural therapeutic potential. *Objective*: To investigate the effects of cinnamon extract in a model of hypercholesterolemia using mice. *Methods*: This study used young male C57BJ6 mice (90 days old) with and without hypercholesterolemia caused by a genetic deficiency in LDL receptors. The animals were given cinnamon extract or vehicle for 30 days. After treatment, blood levels of total cholesterol, LDL, HDL, oxidized LDL and triglycerides were assessed. In addition, markers of oxidative stress in the blood were analyzed, such as total antioxidant capacity (TRAP), levels of reduced glutathione (GSH) and reactive oxygen species (ROS), as well as cytokines related to inflammation in the hippocampus (tumor necrosis factor-α and interleukins 1-β, 6 and 10). *Results*: Mice deficient in the LDL receptor showed increased levels of total cholesterol, LDL, oxidized LDL and triglycerides, as well as reduced HDL. Treatment with cinnamon extract normalized these biochemical parameters, protected against oxidative damage and reduced the production of pro-inflammatory cytokines, increasing anti-inflammatory cytokines in the hippocampus. *Conclusion*: *Cinnamomum zeylanicum* extract showed beneficial effects in the treatment of hypercholesterolemia and may be useful as a preventive or adjuvant therapy.

Keywords: Herbal medicine. Cholesterol. Oxidative stress. Inflammation.

Resumo

Introdução: A hipercolesterolemia constitui o principal fator de risco para a formação de placas ateromatosas nas artérias e é um importante fator de risco para o desenvolvimento da doença de Alzheimer. Dentre as plantas que têm sido alvo de diversos estudos farmacológicos, a canela (*Cinnamomum zeylanicum*) desponta como detentora de um potencial terapêutico natural.

Objetivo: Investigar os efeitos do extrato de canela em um modelo de hipercolesterolemia utilizando camundongos. *Métodos*: Para o presente estudo, foram utilizados camundongos da linhagem C57BJ6, machos jovens (90 dias de idade), com e sem hipercolesterolemia causada por deficiência genética nos receptores de LDL. Os animais receberam, durante 30 dias, extrato de canela ou veículo. Após o tratamento, foram avaliados os níveis sanguíneos de colesterol total, LDL, HDL, LDL oxidada e triglicerídeos. Além disso, foram analisados marcadores de estresse oxidativo no sangue, como capacidade antioxidante total (TRAP), níveis de glutationa (GSH) reduzida e espécies reativas de oxigênio (ROS), além de citocinas relacionadas com inflamação no hipocampo (fator de necrose tumoral-α e interleucinas 1-β, 6 e 10). *Resultados*: Os camundongos com deficiência no receptor de LDL apresentaram aumento dos níveis de colesterol total, LDL, LDL oxidada e triglicerídeos, além de redução do HDL. O tratamento com extrato de canela normalizou esses parâmetros bioquímicos, protegeu contra danos oxidativos e reduziu a produção de citocinas pró-inflamatórias, aumentando as citocinas anti-inflamatórias no hipocampo. *Conclusão*: O extrato de *Cinnamomum zeylanicum* demonstrou efeitos benéficos no tratamento da hipercolesterolemia e pode ser útil como terapia preventiva ou adjuvante.

Palavras-chave: Fitoterápico. Colesterol. Estresse Oxidativo. Inflamação.

INTRODUÇÃO

Cardiovascular diseases (CVD), such as stroke and myocardial infarction, are currently the leading cause of death in Western countries.[1] They originate mainly from complications of atherosclerosis, a syndrome characterized by the accumulation of lipids and inflammatory cells in the inner walls of arteries and arterioles, causing a reduction in blood perfusion to vital organs.[2] Although hypercholesterolemia has traditionally been considered one of the main causes of atherogenesis, it has been widely demonstrated that inflammation and oxidative stress also play key roles in both the progression of atherosclerosis and the formation of vulnerable plaques.[3,4]

Excessive calorie and fat intake is associated with metabolic syndrome changes in lipid and glucose metabolism. High-fat diets, especially those rich in saturated fatty acids, stimulate adipose tissue to produce cytokines, which characterize subclinical inflammation, and also increase the storage of triglycerides in adipocytes.[5] When storage is saturated by excess nutrients, there is a systemic release of free fatty acids and more inflammatory cytokines, which can damage peripheral tissues such as skeletal muscle, liver, heart and pancreas, causing peripheral lipotoxicity, which is an important mechanism for the emergence of insulin resistance.[6]

For hypercholesterolemia, it is recommended to reduce the intake of foods rich in cholesterol and saturated fatty acids. This means restricting the consumption of animal fats such as red meat, whole milk and its derivatives, butter, egg yolks, sausages, animal skin and offal, seafood, creamy ice cream, palm oil and coconut oil. The consumption of soluble fibers represented by pectin (fruits) and gums (legumes, oats and barley) act by delaying gastric emptying and increasing intestinal transit time, slowing down the absorption of glucose, delaying the hydrolysis of starch and reducing the concentrations of total cholesterol and LDL-c, so consumption should be encouraged.[7,8]

Mice with gene deletion for low-density lipoprotein receptors ($LDLr^{-/-}$) are a model of human familial hypercholesterolemia. These animals show hypercholesterolemia, characterized by increased levels of LDL-cholesterol, even when submitted to a standard diet, and may develop atherosclerotic lesions in the long term. Due to the absence of LDL receptors, LDL remains free in the circulation for longer, causing blood cholesterol levels to rise to twice the normal level.[9] These *knockout* animals have been widely used to study the biological mechanisms underlying cardiovascular diseases, as well as to study substances with potential therapeutic effects. In this sense, the search for drugs that can act in the prevention and treatment of factors associated with hypercholesterolemia is a challenge for the pharmaceutical industry and government health agencies.[9]

Even with the development of synthetic drugs, medicinal plants remain an alternative form of treatment in various parts of the world, and in recent decades there has been an increase in the use of herbal preparations for therapeutic purposes.[10] Among the plants that have been the subject of various pharmacological studies, *Cinnamomum zeylanicum* stands out as having therapeutic potential. It is popularly known as cinnamon and is one of the best-known spices used in Portuguese cuisine and in other parts of the world to add flavor, aroma and color to foods and drinks.[11] In addition to its organoleptic characteristics, the polyphenols found in cinnamon may lead to improvements in the components of metabolic syndrome (MS) and a reduction in the risk of factors associated with diabetes and cardiovascular disease. Animal and human studies involving individuals with MetS, type 2 diabetes mellitus (DM) and polycystic ovary syndrome have demonstrated beneficial effects using cinnamon and aqueous extracts of cinnamon with regard to biochemical parameters such as glucose, insulin, lipids and antioxidants, as well as effects on lean body mass and inflammatory response.[12]

Epidemiological and experimental evidence supports the association between alterations in cholesterol metabolism and the onset of cognitive impairment and dementia. Many of the classic vascular risk factors, including hypertension, diabetes mellitus and, in particular, hypercholesterolemia, are also considered risk factors for neurodegenerative diseases, especially Alzheimer's disease.[13] However, the molecular mechanisms by which cholesterol levels contribute to the pathophysiology of neurodegenerative diseases have yet to be fully elucidated. Furthermore, as far as we know, there is no data in the literature on the therapeutic effect of *Cinnamomum zeylanicum* in a transgenic model of hypercholesterolemic mice.

Therefore, the aim of this study was to investigate the therapeutic effects of cinnamon extract on biochemical and neurochemical parameters in a mouse model of hypercholesterolemia.

METHODS

Wild-type male C57BJ6 mice (90 days old) with gene deletion for low-density lipoprotein receptors ($LDLr^{-/-}$) weighing between 40-50g were used. The animals were kept in polypropylene boxes under controlled conditions, with a 12-hour light/dark cycle and temperature (22 ± 2°C). The experiments were conducted in accordance with the principles and procedures described by the Brazilian College of Animal Experimentation (COBEA).

Cinnamon extract (CE) was prepared using dehydrated cinnamon bark (*Cinnamomum zeylanicum)* obtained from Santos Flora Comércio de Ervas Ltda, São Paulo-SP, duly identified and packaged. The dehydrated barks were ground and then the ethanolic extract was prepared, as indicated in the Brazilian Homeopathic Pharmacopoeia (1997). The reagents for determining biochemical parameters were obtained from Labtest Ltda. (Brazil) and the ELISA *kits* for determining inflammatory parameters were obtained from R&D Systems (Minneapolis, MN, USA).

Thirty-six mice were used for all the experiments proposed in this study. The animals were randomly divided into six groups (n= 6/group), three corresponding to the wild-type group and three to the $LDLr^{-/-}$ group: (1) Wild-type/control group; (2) Wild-type/ethanol group; (3) Wild-type/CE group; (4) $LDLr^{-/-}$/control group; (5) $LDLr^{-/-}$/ethanol group; (6) $LDLr^{-/-}$/CE group.The mice were given pure water (control groups), cinnamon ethanolic extract (CE groups) or 70% ethanol vehicle (ethanol group) in their drinking water at a concentration of 4.5 ml/kg/body weight/day for 30 days. The amount of EC was based on the study by Hagenlocher et al.[14] and corresponds to 0.8g of *cinnamomum bark/kg* body weight. The extract was prepared weekly, bottles were changed and the extract intake was monitored. 24 hours after the end of the 30-day treatment with cinnamon extract or vehicle, all the animals were euthanized with an injection of pentobarbital (180mg/kg, intraperitoneally) and the blood and hippocampus were removed for biochemical determinations.

The hippocampus was homogenized in 10Mm Tric-HCl (pH 7.4). The homogenate was centrifuged at 2,400g for 15 minutes at 4ºC and the supernatant fraction (S_1) was used for neurochemical assays. Blood samples were collected by cardiac puncture from the anesthetized animals, using heparin as an anticoagulant, and the plasma was separated by centrifugation (2,400g for 15 minutes).

The biochemical parameters total cholesterol, HDL cholesterol (high density lipoprotein), LDL (low density lipoprotein), LDLox (oxidized LDL), triglycerides, total lipids and VLDL (very low density lipoprotein) were analyzed spectrophotometrically using standardized diagnostic *kits* (Labtest®).The total antioxidant capacity (TRAP) of hippocampal tissue was measured according to the method of Lissi et al.[15] The total antioxidant reactivity (TAR) of the hippocampus was determined by measuring the luminol

chemiluminescence intensity induced by 2,2'azo-bis-(2-amidinopropane) (ABAP) at room temperature.[16] The levels of reactive oxygen species (ROS) in the hippocampus were determined by a spectro-fluorimetric method using the DCHF-DA assay.[17]

Glutathione (GSH) levels in the hippocampus were determined fluorometrically according to Hissin & Hilf.[18] Levels of tumor necrosis factor-alpha (TNF-α), interleukin 1-beta (IL-1β), interleukin-6 (IL-6) and interleukin-10 (IL-10) were analyzed in the hippocampus using Rat Cytokine Duo Set ELISA *kits* from R&D Systems (Minneapolis, MN, USA), according to the manufacturer's instructions (protein range 31.25-2,000pg).

Results were presented as mean and standard error of the mean. Comparisons between groups were analyzed using one-way ANOVA, followed by the Newman-Keuls *post hoc* test. A value of $p<0.05$ was considered significant. All tests were carried out using Graph Pad Prism 5.0 *software.*

RESULTS

Triglyceride levels were elevated in the control (250.3 ± 22.7), ethanol (236.6 ± 26.6) and CE (207 ± 15.0) groups of the LDLr$^{-/-}$ strain, compared to the control (101.1 ± 9.1), ethanol (98.5 ± 8.3) and CE (91.0 ± 6.0) groups of the wild-type strain, respectively. However, the LDLr$^{-/-}$ CE group (207 ± 15.0) showed lower triglyceride levels compared to the control group (250.3 ± 22.7) of the same strain (Figure 1A).

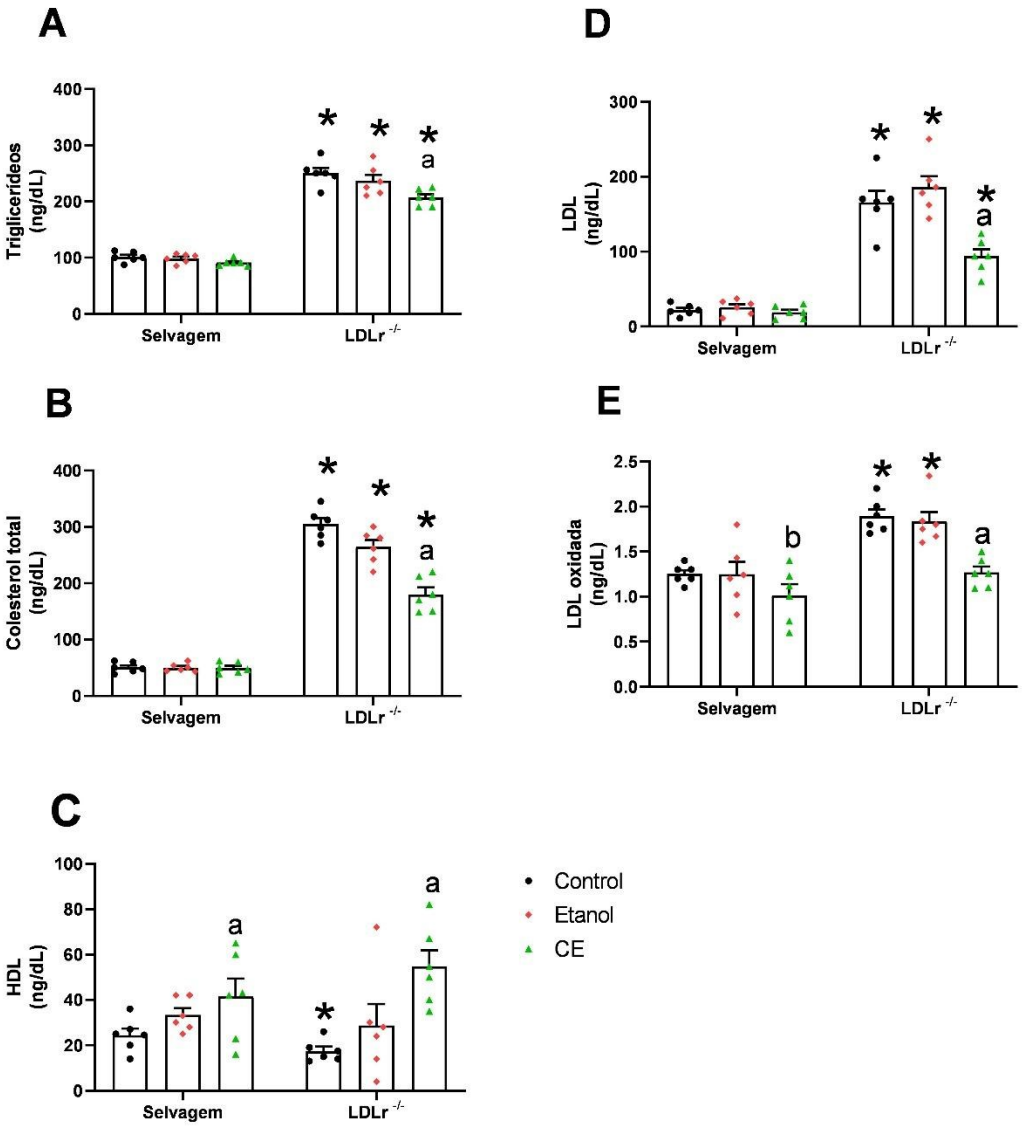
With regard to total cholesterol levels, the LDLr$^{-/-}$ animals in the control (312.1 ± 19.0), ethanol (260 ± 19.3) and CE (179.3 ± 23.7) groups showed higher levels of this marker compared to the control (50.3 ± 9.2), ethanol (49.8 ± 7.2) and CE (49.8 ± 9.6) groups of the wild-type strain. Even so, the LDLr$^{-/-}$ CE group (179.3 ± 23.7) showed lower levels of total cholesterol compared to the control group of the same strain (312.1 ± 19.0) (Figure 1B).

The wild animals in the CE group (41.5 ± 19.4) showed an increase in HDL levels compared to the wild control group (24.4 ± 7.3). Similarly, there was an increase in this parameter in the LDLr$^{-/-}$ CE group (54.7 ± 17.4) compared to the control group of the same strain (17.4 ± 4.7). Finally, the animals in the LDLr$^{-/-}$ control group (17.4 ± 4.7) had lower HDL levels compared to the wild-type control group (24.4 ± 7.3) (Figure 1C).

LDL levels were higher in the control (165.5 ± 38.2), ethanol (185.8 ± 36.7) and CE (93.9 ± 22.6) groups of the LDLr$^{-/-}$ strain, compared to the control (21.8 ± 7.5), ethanol (25.5 ± 8.5) and CE (18.9 ± 8.5) groups of the wild-type strain, respectively. However, the LDLr$^{-/-}$ CE group (93.9 ± 22.6) showed reduced LDL levels compared to the control group (165.5 ± 38.2) of the same strain (Figure 1D).

As for oxidized LDL levels, the LDLr$^{-/-}$ animals in the control (1.8 ± 0.18) and ethanol (1.8 ± 0.26) groups showed higher levels compared to the respective wild-type groups: control (1.2 ± 0.10) and ethanol (1.2 ± 0.34). Wild animals in the CE group (1.01 ± 0.30) showed a reduction in oxidized LDL levels compared to the ethanol group (1.2 ± 0.34), from the same strain. In addition, the LDLr$^{-/-}$ CE group (1.26 ± 0.16) showed a reduction in this marker compared to the control group of the same strain (1.8 ± 0.18) (Figure 1E).

Figure 1: Effect of EC treatment on the levels of (A) triglycerides, (B) total cholesterol, (C) HDL cholesterol, (D) LDL cholesterol and (E) oxidized LDL in the plasma of wild-type and LDLr$^{-/-}$ mice. Data are expressed as mean and standard error of the mean (n=6 per group). * indicates a significant difference when compared to the wild-type group in the same treatment; 'a' and 'b' denote a significant difference when compared to the control group and the ethanol group, respectively, within the same strain (wild-type or LDLr$^{-/-}$). One-way ANOVA was performed followed by the Newman-Keuls *post hoc* multiple comparisons test.



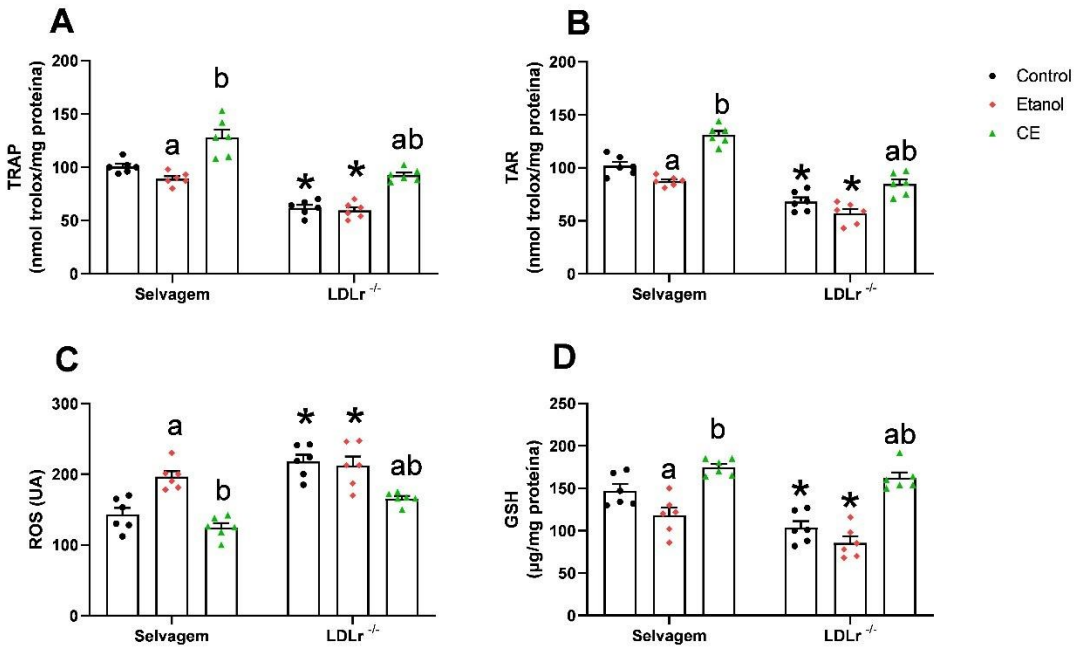
The post-hoc test showed a reduction in TRAP levels in the ethanol group (92.8 ± 6.1), compared to the control (100 ± 6.2), and an increase in the CE group (128.2 ± 17.5) compared to the ethanol group, in the wild-type strain. In the LDLr$^{-/-}$ strain, the CE group (92.5 ± 5.8) showed an increase in TRAP levels compared to the control (64.3 ± 11.7) and ethanol (62.3 ± 17.7) groups. Finally, the animals in the control and ethanol groups of the LDLr$^{-/-}$ strain showed reduced levels of TRAP compared to the respective groups of the wild-type strain (Figure 2A).

As for TAR levels, there was a reduction in the ethanol group (87.3 ± 4.5) compared to the control (100 ± 18.0) and an increase in the CE group (131.3 ± 8.9) compared to ethanol in the wild-type strain. In the LDLr$^{-/-}$ strain, the CE group (84.0 ± 10.4) showed an increase in TAR levels compared to the control (68.2 ± 9.3) and ethanol (57.0 ± 9.9) groups. Comparing the strains, the control and ethanol groups of the LDLr$^{-/-}$ strain showed reduced levels of TAR compared to the respective groups of the wild-type strain (Figure 2B).

In the assessment of ROS generation, wild-type animals in the ethanol group (182.5 ± 30.7) showed an increase in this marker compared to the control (153.6 ± 37.9) and CE groups (124.4 ± 22.5). In the LDLr$^{-/-}$ strain, the CE group (165.8 ± 8.6) showed a reduction in ROS levels compared to the control (218.4 ± 22.5) and ethanol (212.5 ± 20.8) groups. In addition, the control and ethanol groups of the LDLr$^{-/-}$strain showed higher values than the respective groups of the wild-type strain (Figure 2C).

There was a reduction in GSH levels in the ethanol group (118.3 ± 22.2) compared to the control (147.1 ± 18.7), and an increase in the CE group (184.6 ± 12.4) compared to ethanol, in the wild-type strain. In the LDLr$^{-/-}$ strain, the CE group (152.3 ± 29.8) showed increased GSH levels compared to the control (103.6 ± 18.4) and ethanol (85.8 ± 18.3) groups. Finally, the control and ethanol groups of the LDLr$^{-/-}$ strain showed a reduction in GSH compared to the same groups of the wild-type strain (Figure 2D).

Figure 2: Effect of EC treatment on the levels of (A) TRAP, (B) TAR, (C) ROS and (D) GSH in the hippocampus of wild-type and LDLr$^{-/-}$ mice. Data are expressed as mean and standard error of the mean (n=6 per group). * indicates a significant difference when compared to the wild-type group in the same treatment; 'a' and 'b' denote a significant difference when compared to the control group and the ethanol group, respectively, within the same strain (wild-type or LDLr$^{-/-}$). One-way ANOVA was performed followed by the Newman-Keuls *post hoc* multiple comparisons test.



In the wild-type strain, the CE group (19.8 ± 8.6) showed a reduction in TNF-α levels compared to the ethanol group (42.4 ± 5.7). In the LDLr$^{-/-}$ strain, the CE group (75.1 ± 20.7) showed a reduction in this marker compared to the control (145.4 ± 28.3) and ethanol (175.8 ± 6.2) groups. In addition, the control and ethanol

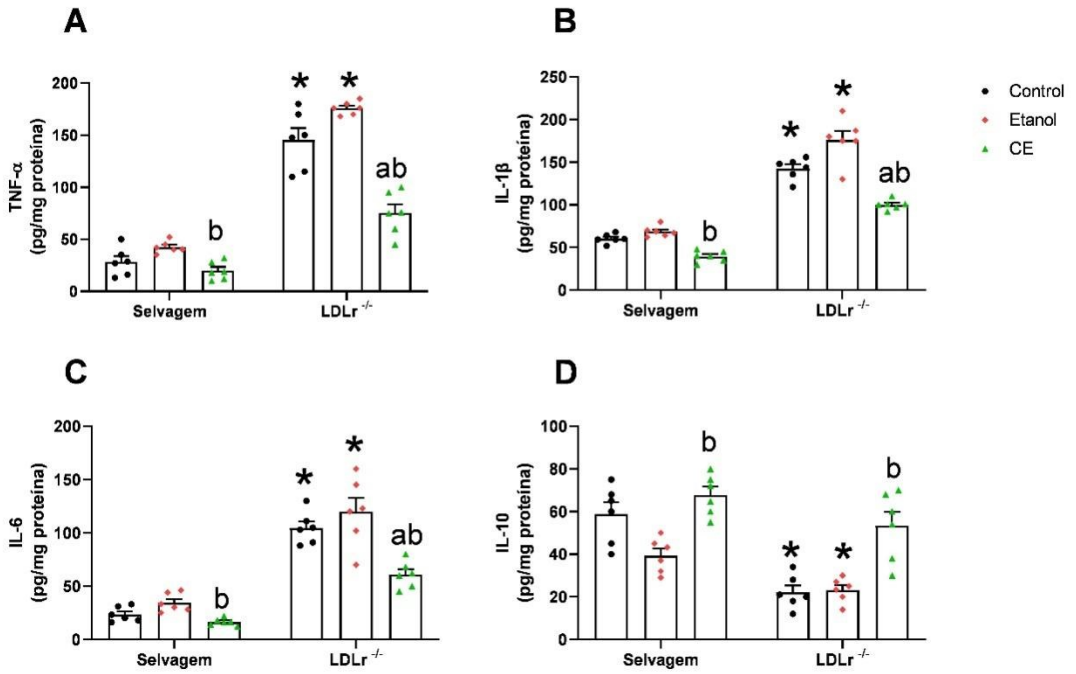
groups of the LDLr$^{-/-}$ strain showed increased levels of TNF-α compared to the respective groups of the wild-type strain (Figure 3A).

Wild-type animals in the CE group (39.7 ± 6.5) showed reduced IL-1β levels compared to the ethanol group (68.62 ± 6.3). In the LDLr$^{-/-}$ strain, the CE group (100.1 ± 5.9) showed a reduction compared to the control (142.4 ± 12.4) and ethanol (176.1 ± 26.1) groups. The control and ethanol groups of the LDLr$^{-/-}$ strain also showed increased levels of IL-1β compared to the respective groups of the wild-type strain (Figure 3B).

In the wild-type strain, the CE group (16.4 ± 3.4) showed a reduction in IL-6 levels compared to the ethanol group (34.3 ± 8.6). In the LDLr$^{-/-}$ strain, the CE group (60.8 ± 12.5) showed a reduction in levels compared to the control (104.6 ± 15.1) and ethanol (120 ± 31.8) groups. In addition, the control and ethanol groups of the LDLr$^{-/-}$ strain showed higher levels of IL-6 compared to the respective groups of the wild-type strain (Figure 3C).

Animals in the EC group (66.1 ± 11.7) of the wild-type strain showed increased levels of IL-10 compared to the ethanol group (42.7 ± 12.5). Similarly, animals in the LDLr$^{-/-}$ CE group (53.33 ± 16.2) showed higher levels compared to the ethanol group (23.2 ± 5.6) of the same strain. Finally, the control (21.5 ± 6.6) and ethanol (23.2 ± 5.6) groups of the LDLr$^{-/-}$ strain showed reduced levels of this marker compared to the respective groups of the wild-type strain (Figure 3D).

Figure 3: Effect of EC treatment on the levels of (A) TNF-α, (B) Il-1β, (C) IL-6 and (D) IL-10 in the hippocampus of wild-type and LDLr$^{-/-}$ mice. Data are expressed as mean and standard error of the mean (n=6 per group). * indicates a significant difference when compared to the wild-type group in the same treatment; 'a' and 'b' denote a significant difference when compared to the control group and the ethanol group, respectively, within the same strain (wild-type or LDLr$^{-/-}$). One-way ANOVA was performed followed by the Newman-Keuls *post hoc* multiple comparisons test.



DISCUSSION

Atherosclerosis is an inflammatory disease characterized by the formation of plaques inside the arteries, causing them to thicken and lose elasticity. These plaques result from the accumulation of lipids and cholesterol. When atherosclerosis forms, blood is prevented from circulating normally, leading to the development of myocardial ischemia. The development of atherosclerosis causes the onset of cardiovascular diseases that can lead to death, such as infarction and/or heart attack. Atherosclerosis is one of the main causes of death from cardiovascular disease.[19,20] The development of this disease results from high concentrations of total cholesterol, triglycerides, low-density lipoprotein (LDL) cholesterol and/or a low level of high-density lipoprotein (HDL) cholesterol. LDL is oxidized through the action of free radicals or by enzymes that mediate its oxidation. Once altered, LDL binds to the artery walls, forming fatty plaques, thus initiating the inflammatory process.[20,21] Plasma high-density lipoprotein (HDL) promotes the outflow of cholesterol, which is inversely associated with the risk of atherosclerotic CVD.[22]

The present study showed that LDLr$^{-/-}$ animals had increased levels of TG, TC, LDL and LDLox and decreased levels of HDL cholesterol. Cinnamon extract prevented these changes in biochemical parameters caused by the absence of the LDL receptor. In another study, cinnamon and its extract showed lipid-lowering effects in animal experiments. Treatment with cinnamic acid, a phytochemical compound found naturally in various plants, especially in essential oils extracted from cinnamon bark and leaves and with many biological effects, reduced TC and LDL-C levels and increased high-density lipoprotein cholesterol (HDL-C) levels in mice.[23] In addition, the aqueous extract of *C. zelanicum* significantly prevented dyslipidemia and protected the aortas from atherosclerotic changes induced by dexamethasone in Wistar rats.[24]

Favorable results have also been found in humans further east on the Asian continent, specifically in India. Researchers have shown that continuous and prolonged use of cinnamon reduced not only LDL, but also TG and TC values.[25] Another study showed that treatment of animals for a period of thirty days with an aqueous extract of cinnamon is active against DM (diabetes mellitus) since it influences glucose concentration, reduces food consumption and lowers cholesterol levels.[26] Similarly, another study showed the long-lasting antihypertensive effect of administering cinnamon extract (5, 10 and 20 mg/Kg) in a rat model, as well as significantly reducing levels of triglycerides, total cholesterol, LDL-cholesterol and increasing HDL-cholesterol in plasma.[27] A review study on medicinal plants that can be used as functional foods reported that the consumption of some species of cinnamon may have beneficial effects in the prevention and management of cardiovascular diseases, hypertension and diabetes, due to its antioxidant properties.[28]

There is great interest in antioxidant studies, mainly due to discoveries about the effect of free radicals on the body.[29] Free radicals and other reactive oxygen species (ROS) are toxic molecules and can cause damage to DNA, membranes and cell death in extreme cases.[29] In this context, oxidative stress is related to the pathogenic mechanisms of atherosclerosis and neurodegenerative diseases.

In the present study, the antioxidant potential of cinnamon extract was assessed using the TRAP technique, which measures the time that the sample or standard maintains free radical sequestration, and the TAR index, which reflects the capacity and speed of a substance or mixture of them to participate in the process of electron transfer to luminol-derived radicals,[30,31] in other words, it measures the capacity to react in the face of an increase in free radicals. In this respect, the present study showed that cinnamon extract reversed the decrease in TAR and TRAP levels in the hippocampus of hypercholesterolemic animals, corroborating previous studies which have shown that cinnamon extract has defense mechanisms against cell damage caused by free radicals in order to maintain cellular balance.[32]

Among the mammalian antioxidant defense systems, GSH (glutathione), a low molecular weight tripeptide composed of glutamate, cysteine and glycine, is considered the main endogenous non-enzymatic antioxidant. Among its antioxidant functions, GSH acts as a chelator of free radicals and reactive oxygen species.[33] In the present study, we demonstrated that cinnamon extract was able to reverse GSH depletion in the hippocampus of hypercholesterolemic animals. A probable mechanism for the antioxidant effect of cinnamon is the presence of cinnamaldehyde (an aromatic acid naturally found in cinnamon oil (*Cinnamomum zeylanicum)*, which is capable of raising glutathione levels.[34] In addition, Khedkar et al.[35] concluded that cinnamon extract has significant antioxidant activity in humans. Therefore, cinnamon, which is used as a flavoring agent in food or tea, can act as a potent antioxidant and can be used in individuals who have diseases related to oxidative stress.

Despite the impact that cardiovascular diseases have, they can be controlled by adopting healthy lifestyle habits, as well as correcting some of the risk factors associated with them, such as hypercholesterolemia, hypertension and oxidative stress.[36]

It has been suggested that hypercholesterolemia can induce alterations in the blood-brain barrier, causing extravasation of serum components into the brain through the wall of small cerebral vessels, allowing a recruitment of immune-inflammatory cells from the bloodstream.[13] These deleterious effects may be related to neuroinflammation and oxidative damage in brain structures in response to hypercholesterolemia, increasing the risk of developing Alzheimer's disease.

In the present study, we found that LDLr$^{-/-}$ animals had markedly elevated levels of the inflammatory cytokines TNF-α, IL-1β and IL-6, as well as decreased levels of the anti-inflammatory cytokine IL-10 in the hippocampus. This corroborated previous studies in which the polyphenolic fraction of cinnamon showed anti-inflammatory and anti-arthritic activity in animal models at doses of 200 mg/kg, with a high reduction in tumor necrosis factors (TNF-α).[37] He also confirmed that daily consumption of proanthocyanidins - a phenolic compound found in cinnamon leaves - helps prevent low-grade inflammation, both systemic and local, in adipose tissue, as well as in muscle and liver, which improves obesity-induced insulin resistance.[38]

CONCLUSION

The study showed that *Cinnamomum zeylanicum* (cinnamon) extract has significant therapeutic effects in the treatment of hypercholesterolemia, a condition associated with cardiovascular disease and possibly Alzheimer's disease. After 30 days of treatment with cinnamon extract, the biochemical changes in the treated animals were normalized, with a reduction in cholesterol and triglyceride levels and an increase in HDL.

Cinnamon is therefore a natural and effective alternative for controlling high cholesterol. In addition, the treatment showed antioxidant properties. These results obtained in animals indicate that cinnamon extract can be useful not only for prevention, but also in the adjuvant therapy of pathologies.

REFERENCES

1. Roth GA, Mensah GA, Johnson CO, Addolorato G, Ammirati E, Baddour LM, et al. Global burden of cardiovascular diseases and risk factors, 1990–2019: update from the GBD 2019 study. J Am Coll Cardiol. 2020;76(25):2982–3021. https://doi.org/10.1016/j.jacc.2020.11.010

2. Poyatos P, Gratacós M, Samuel K, Orriols R, Tura-Ceide O. Oxidative stress and antioxidant therapy in pulmonary hypertension. Antioxidants (Basel). 2023;12(5):1006. https://doi.org/10.3390/antiox12051006

3. Kong P, Cui ZY, Huang XF, Zhang DD, Guo RJ, Han M. Inflammation and atherosclerosis: signaling pathways and therapeutic intervention. Signal Transduct Target Ther. 2022;7(1):131. https://doi.org/10.1038/s41392-022-00955-7

4. Raggi P, Genest J, Giles JT, Rayner KJ, Dwivedi G, Beanlands RS, et al. Role of inflammation in the pathogenesis of atherosclerosis and therapeutic interventions. Atherosclerosis. 2018;276:98–108. https://doi.org/10.1016/j.atherosclerosis.2018.07.014

5. Denisenko YK, Kytikova OY, Novgorodtseva TP, Antonyuk MV, Gvozdenko TA, Kantur TA. Lipid-induced mechanisms of metabolic syndrome. J Obes. 2020;2020:5762395.https://doi.org/10.1155/2020/5762395

6. Kojta I, Chacińska M, Błachnio-Zabielska A. Obesity, bioactive lipids, and adipose tissue inflammation in insulin resistance. Nutrients. 2020;12(5):1305. https://doi.org/10.3390/nu12051305

7. Ahmad N, Shabbir U, Sameen A, Manzoor MF, Ahmad MH, Ismail T, et al. Hypocholesterolemic effect of designer yogurts fortified with omega fatty acids and dietary fibers in hypercholesterolemic subjects. Food Sci Technol. 2021;41:1000–8. https://doi.org/10.1590/fst.22420

8. Cunha AFC, Ribeiro I. Hipercolesterolemia familiar: a importância do diagnóstico e tratamento precoces. Int J Cardiovasc Sci. 2017;30:550–3. https://doi.org/10.5935/2359-4802.20170077

9. Andreadou I, Schulz R, Papapetropoulos A, Ferdinandy P, Hausenloy DJ, Daiber A, et al. Hyperlipidaemia and cardioprotection: animal models for translational studies. Br J Pharmacol. 2020;177(23):5287–311. https://doi.org/10.1111/bph.14931

10. Dewi MK, Zahra A, Saadah M, Hidayat AT, Pramono S, Nugraha AP, et al. Improved activity of herbal medicines through nanotechnology. Nanomaterials. 2022;12(22):4073. https://doi.org/10.3390/nano12224073

11. Hussain Z, Khan JA, Rashid H. Cinnamomum zeylanicum (Darchini): a boon to medical science and a possible therapy for stress-induced ailments. Crit Rev Eukaryot Gene Expr. 2019;29(3):277–86. https://doi.org/10.1615/CritRevEukaryotGeneExpr.2019028867

12. Beji RS, Nauli SM, El-Sherbeni AA, Pathak DR, Chen H. Antidiabetic, antihyperlipidemic and antioxidant influences of the spice cinnamon (Cinnamomum zeylanicum) in experimental rats. Braz J Pharm Sci. 2018;54:e17576. https://doi.org/10.1590/s2175-97902018000217576

13. De Bem AF, da Silva FP, Retamoso LT, Dornelles GL, da Rocha JBT, da Silva LGG, et al. Animal models of metabolic disorders in the study of neurodegenerative diseases: an overview. Front Neurosci. 2021;14:604150. https://doi.org/10.3389/fnins.2020.604150

14. Hagenlocher Y, Feilhauer K, Schäfer KH, Kleinschnitz C, Geißlinger G, Lührs H, et al. Cinnamon extract reduces symptoms, inflammatory mediators and mast cell markers in murine IL-10(−/−) colitis. J Nutr Biochem. 2016;30:85–92. https://doi.org/10.1016/j.jnutbio.2015.11.015

15. Lissi E, Salim-Hanna M, Pascual C, del Castillo MD. Evaluation of total antioxidant potential (TRAP) and total antioxidant reactivity from luminol-enhanced chemiluminescence measurements. Free Radic Biol Med. 1995;18:153–8. https://doi.org/10.1016/0891-5849(94)00117-3

16. Lissi E, Pascual C, del Castillo MD. Luminol luminescence induced by 2,2'-Azo-bis(2-amidinopropane) thermolysis. Free Radic Res Commun. 1992;17:299–311. https://doi.org/10.3109/10715769209079523

17. Loetchutinat C, Drouin G, Pradines A, Allal C, Bagrel D. Spectrofluorometric determination of intracellular levels of reactive oxygen species in drug-sensitive and drug-resistant cancer cells using the 2′,7′-dichlorofluorescein diacetate assay. Radiat Phys Chem. 2005;72:323–31. https://doi.org/10.1016/j.radphyschem.2004.06.011

18. Hissin PJ, Hilf R. A fluorometric method for determination of oxidized and reduced glutathione in tissues. Anal Biochem. 1976;74:214–26. https://doi.org/10.1016/0003-2697(76)90326-2

19. Jebari-Benslaiman S, Galicia-Garcia U, Larrea-Sebal A, Olaetxea JR, Alloza I, Varela-Rey M, et al. Pathophysiology of atherosclerosis. Int J Mol Sci. 2022;23(6):3346. https://doi.org/10.3390/ijms23063346

20. Frąk W, Saeed S, Hegazy GA, Bil-Lula I, Witkiewicz W, Rybak Z, et al. Pathophysiology of cardiovascular diseases: new insights into molecular mechanisms of atherosclerosis, arterial hypertension, and coronary arterydisease.Biomedicines.2022;10(8):1938. https://doi.org/10.3390/biomedicines10081938

21. Ito F, Ito T. High-density lipoprotein (HDL) triglyceride and oxidized HDL: new lipid biomarkers of lipoprotein-related atherosclerotic cardiovascular disease.Antioxidants.2020;9(5):362. https://doi.org/10.3390/antiox9050362

22. Ouimet M, Barrett TJ, Fisher EA. HDL e transporte reverso de colesterol: mecanismos básicos e seus papéis na saúde e doença vascular. *Circ Res*.2019;124(10):1505-18. https://doi.org/10.1161/CIRCRESAHA.118.313086

23. Shang C, Lin H, Fang X, Wang Y, Jiang Z, Qu Y, et al. Efeitos benéficos da canela e seus extratos no tratamento de doenças cardiovasculares e diabetes. Food Funct. 2021;12(24):12194–12220. https://doi.org/10.1039/D1FO01935J

24. Nayak IN, Chinta R, Jetti R. Potencial antiaterosclerótico do extrato aquoso da casca de Cinnamomum zeylanicum contra aterosclerose induzida por glicocorticoides em ratos Wistar. J Clin Diagn Res. 2017;11:FC19–FC23. https://doi.org/10.7860/JCDR/2017/23910.9864

25. Gupta JS, Gupta P, Singh A, Mishra R, Tripathi P, Dubey D, et al. Effect of oral cinnamon intervention on metabolic profile and body composition of Asian Indians with metabolic syndrome: a randomized double-blind control trial. Lipids Health Dis. 2017;16:113. https://doi.org/10.1186/s12944-017-0504-8

26. Lira JCG, Lima GPV, Vitorino DF, Silva ML, Almeida JMC, Araujo ASR, et al.Effectiveness of cinnamon in the reduction of lipid levels in people with diabetes: a randomized clinical trial. Rev Gaúcha Enferm. 2023;44:e20230051. https://doi.org/10.1590/19831447.2023.20230051.n

27. Nyadjeu P, Nguelefack-Mbuyo EP, Atsamo AD, Nguelefack TB, Dongmo AB, Kamanyi A. Acute and chronic antihypertensive effects of Cinnamomum zeylanicum stem bark methanol extract in L-NAME-induced hypertensive rats. BMC Complement Altern Med. 2013;13:27. https://doi.org/10.1186/1472-6882-13-27

28. Das G, Gonçalves S, Heredia JB, Romano A, Jiménez-Ortega LA, Gutiérrez-Grijalva EP, et al. Cardiovascular protective effect of cinnamon and its major bioactive constituents: an update. J Funct Foods. 2022;97:105045. https://doi.org/10.1016/j.jff.2022.105045

29. Fang YZ, Yang S, Wu G. Free radicals, antioxidants, and nutrition. Nutrition. 2002;18(10):872–9. https://doi.org/10.1016/s0899-9007(02)00916-4

30. Shahid MZ, Butt MS, Sultan MT, Shahid M, Minhas R, Ranjha MMAN. Antioxidant capacity of cinnamon extract for palm oil stability. Lipids Health Dis. 2018;17:1–8. https://doi.org/10.1186/s12944-018-0756-y

31. Munteanu IG, Apetrei C. Analytical methods used in determining antioxidant activity: a review. Int J Mol Sci. 2021;22(7):3380. https://doi.org/10.3390/ijms22073380

32. Isik B, Aydin F, Yilmaz N, Daglioglu N, Yildirim B, Altinkaya SO, et al. Protective effect of cinnamon extract against cobalt-induced multiple organ damage in rats. Front Pharmacol. 2024;15:1384181. https://doi.org/10.3389/fphar.2024.1384181

33. Farina M, Aschner M. Glutathione antioxidant system and methylmercury-induced neurotoxicity: an intriguing interplay. Biochim Biophys Acta Gen Subj. 2019;1863(12):129285. https://doi.org/10.1016/j.bbagen.2019.01.007

34. Pagliari S, Petrillo F, Pacioni G, Viscogliosi G, Modesti A, Pinto A, et al. Antioxidant and anti-inflammatory effect of cinnamon (Cinnamomum verum J. Presl) bark extract after in vitro digestion simulation. Foods. 2023;12(3):452. https://doi.org/10.3390/foods12030452

35. Khedkar S, Khan MA. Aqueous extract of cinnamon (Cinnamomum spp.): role in cancer and inflammation. Evid Based Complement Alternat Med. 2023;2023:5467342. https://doi.org/10.1155/2023/5467342

36. Ghodeshwar GK, Dube A, Khobragade D. Impact of lifestyle modifications on cardiovascular health: a narrative review. Cureus. 2023;15(7):e42616. https://doi.org/10.7759/cureus.42616

37. Inoue Y, Akimoto T, Matsuoka Y, Fukunaga M, Tanaka M, Nakagawa T, et al. Pathophysiology and probable etiology of cerebral small vessel disease in vascular dementia and Alzheimer's disease. Mol Neurodegener. 2023;18(1):46. https://doi.org/10.1186/s13024-023-00640-5

38. Sayed UFSM, Ghori SS, Choudhury SM, Bhatia S, Swamy KV, Patel K, et al. Natural products as novel anti-obesity agents: insights into mechanisms of action and potential for therapeutic management. Front Pharmacol.2023;14:1182937. https://doi.org/10.3389/fphar.2023.1182937

Contributors

Bálsamo EC participated in the execution of the experiment, analysis and interpretation of data, and writing of the manuscript; Madalosso LM and Bertolazi SB contributed to the revision of the manuscript and organization of bibliographical references; Segat HJ contributed to the revision of the manuscript and analysis and interpretation of data; Boeira SP participated as coordinator of the research project, reviewing and approving the final version.

Conflict of Interest: The authors declare no conflict of interest.

Received: May 20, 2024
Accepted: April 14, 2025