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Original Article

The effect of ethanol-based coriander (*Coriandrum sativum* L.) seed extract on oxidative stress, antioxidant level and cellular senescence in the heart of obese rat

[El efecto del extracto de semilla de cilantro (*Coriandrum sativum* L.) a base de etanol sobre el estrés oxidativo, el nivel de antioxidantes y la senescencia celular en el corazón de ratas obesas]

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Abstract

Context: High-fat diets contribute to oxidative stress and trigger cellular senescence through an imbalance in reactive oxygen species. The potential of coriander (*Coriandrum sativum*) seed, known for its therapeutic properties against oxidative stress and senescence in obese hearts, has been relatively unexplored.

Aims: To evaluate the impact of ethanol-based *C. sativum* seed extract on oxidative stress, antioxidant levels, and senescence parameters in the hearts of obese rats induced by a high-fat diet.

Methods: Twenty-nine male Wistar rats were divided into five groups, fed different diets for 24 weeks, and received *C. sativum* extract treatment for 12 weeks. This experiment assessed malondialdehyde (MDA) as an oxidative stress marker, antioxidant level by measuring catalase and glutathione (GSH), cellular senescence state by measuring senescence-associated β-galactosidase (SA-β-Gal) activity, and p21 levels in heart tissue.

Results: *C. sativum* seed extract demonstrated a significant reduction in MDA levels in the hearts of obese rats when compared to the control groups. Furthermore, the extract led to a significant increase in catalase and GSH levels in the hearts of non-obese rats, whether on a normal or high-fat diet. Although the *C. sativum*-treated groups exhibited a downward trend in senescence markers (SA-β-Gal and p21), the observed differences did not reach statistical significance.

Conclusions: Ethanol-based *C. sativum* seed extract exhibited promising potential in mitigating oxidative stress in the hearts of obese rats and enhancing antioxidant levels in the hearts of non-obese rats subjected to both normal and high-fat diets. This underscores the preventive role of *C. sativum* seed extract in alleviating oxidative stress, particularly in the context of a high-fat diet. However, there was insufficient evidence to conclusively demonstrate a significant improvement in cellular senescence with the use of ethanol-based *C. sativum* seed extract.

Keywords: antioxidant; cellular senescence; coriander; high-fat diet; oxidative stress.

Resumen

Contexto: Las dietas ricas en grasas contribuyen al estrés oxidativo y desencadenan la senescencia celular a través de un desequilibrio en las especies reactivas de oxígeno. El potencial de la semilla de cilantro (*Coriandrum sativum*), conocida por sus propiedades terapéuticas contra el estrés oxidativo y la senescencia en corazones obesos, ha sido relativamente inexplorado.

Objetivos: Evaluar el impacto del extracto de semilla de *C. sativum* a base de etanol sobre el estrés oxidativo, los niveles de antioxidantes y los parámetros de senescencia en los corazones de ratas obesas inducidas por una dieta rica en grasas.

Métodos: Veintinueve ratas Wistar macho se dividieron en cinco grupos, se alimentaron con diferentes dietas durante 24 semanas y recibieron tratamiento con extracto de *C. sativum* durante 12 semanas. Este experimento evaluó el malondialdehído (MDA) como marcador de estrés oxidativo, el nivel de antioxidante midiendo catalasa y glutatión (GSH), el estado de senescencia celular midiendo la actividad de la β-galactosidasa (SA-β-Gal) asociada a la senescencia y los niveles de p21 en el tejido cardíaco.

Resultados: El extracto de semilla de *C. sativum* demostró una reducción significativa en los niveles de MDA en los corazones de ratas obesas en comparación con los grupos de control. Además, el extracto provocó un aumento significativo de los niveles de catalasa y GSH en el corazón de ratas no obesas, ya sea con una dieta normal o rica en grasas. Aunque los grupos tratados con *C. sativum* mostraron una tendencia a la baja en los marcadores de senescencia (SA-β-Gal y p21), las diferencias observadas no alcanzaron significación estadística.

Conclusiones: El extracto de semilla de *C. sativum* a base de etanol mostró un potencial prometedor para mitigar el estrés oxidativo en los corazones de ratas obesas y mejorar los niveles de antioxidantes en los corazones de ratas no obesas sometidas a dietas tanto normales como ricas en grasas. Esto subraya el papel preventivo del extracto de semilla de *C. sativum* para aliviar el estrés oxidativo, particularmente en el contexto de una dieta rica en grasas. Sin embargo, no hubo pruebas suficientes para demostrar de manera concluyente una mejora significativa en la senescencia celular con el uso de extracto de semilla de *C. sativum* a base de etanol.

Palabras Clave: antioxidante; senescencia celular; cilantro; dieta rica en grasas; estrés oxidativo.

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Abbreviations: CVD: cardiovascular disease; HFD: high-fat diet; GSH: glutathione; IUPAC: International Union of Pure and Applied Chemistry; MDA: malondialdehyde; NADPH: nicotinamide adenine dinucleotide phosphate; Nrf2: nuclear factor erythroid 2-related factor 2; PGC1α: peroxisome proliferator-activated receptor-γ coactivator; PKC: protein kinase C; ROS: reactive oxygen species; SA-β-Gal: senescence-associated β-galactosidase; SIRT1: sirtuin 1.

INTRODUCTION

Obesity is closely related to dyslipidemia, which is accompanied by increased triglyceride levels (Santos and Sinha, 2021). Dyslipidemia, especially non-optimal cholesterol levels, is one of the most important risk factors for atherosclerotic cardiovascular disease (CVD), which accounts for the most deaths worldwide (Mohamed-Yassin et al., 2021). Dyslipidemia plays a pivotal role in the cellular senescence of endothelial cells, which damages the endothelial barrier and promotes lipid accumulation, leading to atherosclerosis development (Xiang et al., 2022). The pathogenesis of CVD, such as atherosclerosis, myocardial infarction, and cardiac fibrosis, can be influenced by an accumulation of the senescence of many cardiac cell types (Chen et al., 2022). Several pathways of cellular senescence are related to cardiovascular disorders. One of the important mechanisms that causes heart failure and hypertrophy in end-stage human heart failure is the apoptosis of cardiomyocytes, which is correlated with the pathway that connects with cell senescence (Shimizu and Minamino, 2019).

A high-fat diet (HFD) is associated with oxidative stress that drives cellular senescence (Tan and Norhaizan, 2019), causing an imbalance between reactive oxygen species (ROS) production and the antioxidant system. Free fatty acids, such as palmitate, stimulate ROS production via the protein kinase C (PKC)-dependent activation of NADPH oxidase in smooth muscle cells and endothelial cells (Zhou et al., 2021). In one study, increased free fatty acids resulted in the peroxidation of lipid products, such as malondialdehyde (MDA) (Jiang et al., 2021). MDA damages cellular function by inactivating membrane transporters, causing peroxidation that increases membrane permeability (Ozata et al., 2002).

Specific increases in ROS levels have been demonstrated to be potentially critical for the induction and maintenance of the cell senescence process (Davalli et al., 2016). Protein carbonyls, protein oxidative alterations, lipofuscin, and DNA damage as oxidation products were observed to accumulate in senescent cell populations, along with increased ROS production (Lawless et al., 2012). ROS accumulation contributes to cardiac dysfunction through lipid peroxidation, extracellular matrix remodeling, mitochondrial damage, and alterations in the coupling proteins (Li et al., 2017). One study showed that the period of HFD feeding affected the cardiac muscle; a chronic HFD induces myocardial hypertrophy and fibrosis in 11 months (Wang et al., 2015). In another study, HFD feeding for six months induced myocardial hypertrophy, hyperglycemia, hyperlipidemia, and cardiac hypertrophy in mice (Fang et al., 2008; Hsu et al., 2016). In addition, HFD feeding triggered elevated myocardial apoptosis (Bhandari et al., 2011). In *Drosophila,* an HFD also causes cardiac lipid accumulation and reduced cardiac contractility (Birse et al., 2010).

Coriander (*Coriandrum sativum* L., family *Apiaceae*) is one of the most popular herbs known for its therapeutic use. Phenolic acids, coumarins, flavonoids, carotenoids, tocopherols, fatty acids, sterols, and essential oils are *C. sativum*-containing compounds, and several studies have shown that the cardioprotective efficacy of *C. sativum* seeds includes antioxidant antiarrhythmic properties and improved lipid profiles and cardiac biomarkers (Mahleyuddin et al., 2021). Numerous in vitro studies have demonstrated that the treatment of senescent cells with flavonoids can produce senolytic effects, postpone replicative senescence, protect several cell types against stress-induced senescence, and regenerate senescent cells (Mária and Ingrid, 2017). The chemical constituents of coriander seeds exhibit a variety of properties capable of protecting against oxidative stress and reducing senescence in the heart. *C. sativum* seed extract is promising as an herbal remedy for maintaining heart health and protecting against CVD. However, the role of *C. sativum* seed ethanolic extract in repairing oxidative stress and cellular senescence of the heart in obese conditions has not been studied.

Therefore, this research aims to evaluate the potential therapeutic role of *C. sativum* seed ethanolic extract in combating oxidative stress (by measuring MDA), improving the antioxidant system (by measuring glutathione and catalase), and eliminating cellular senescence (by measuring SA-*β*-Gal, p21) in the hearts of obese rats induced by high-fat diets. Additionally, this research delved into the potential of *C. sativum* in

preventing cellular senescence by reducing oxidative stress.

MATERIAL AND METHODS

Plant collection, extraction, and GC-MS analysis

The *Coriandrum sativum* L. species of the *Apiaceae* family was identified and assigned by Anom Bowolaksono, PhD, Head of the Biology Department of the Faculty of Mathematics and Natural Science at Universitas Indonesia. The voucher specimen (seeds) was deposited at the Herbarium Depokensis, Universitas Indonesia Biota Collection Room, with reference number JI23-P-053. The *C. sativum* seeds were collected from a horticultural garden in Temanggung Regency, Central Java, Indonesia (GPS location: 7°7'0" S, 110°6'0" E).

The coriander seed extract was prepared in the laboratory of the Department of Chemistry, Faculty of Medicine Universitas Indonesia**.** The *C. sativum* seeds were ground into a powder and macerated with ethanol 96% solvent for 24 hours. The filtrate was evaporated at 50°C, and the resulting extract was freeze-dried to create a denser form that was soluble in water and more durable. This process was conducted in the Indonesia Medical Education and Research Institute IMERI Drug Development Research Cluster. The prepared extract was stored in a refrigerator at a temperature of 4°C.

The phytochemical of the extract was analyzed qualitatively by Advanced Characterization Laboratories Serpong, National Research and Innovation Agency through Gas chromatography-mass spectrometry (GC-MS) method (Agilent 7890B GC with 5977A MSD). The instrument was equipped with a (5%-phenyl)-methylpolysoxane phase capillary column (30 m × 250 µm × 0.25 µm dimension; 0–325°C temperature range). The temperature was initiated at 40°C and post-run at 300°C with a 1 µL injection volume. The analysis was also prepared using N,O-bis(trimethylsilyl)trifluoroacetamide(BSTFA) derivatization.

Animal study

This experiment used 29 male Wistar rats who were nine weeks old. As a previous study explained, the rats were divided into five groups according to the diet administered for 24 weeks (Hardiany et al., 2022b), as shown in Fig. 1. The rats were housed in cages (three animals per cage) under standard temperature conditions of 25 ± 2°C, with a 12-hour light-dark cycle. They were fed a standard or high-fat diet and water *ad libitum*. The groups were classified as follows: control normal diet group N (the normal control group was administered a standard diet for a duration of 24 weeks, n = 5), F (the fat control or obese rat group received a high-fat diet for a duration of 24 weeks, n = 6), NE (the normal control group underwent extract treatment for 12 weeks in conjunction with a standard diet maintained for a duration of 24 weeks, n = 6), FE (the fat control or obese rat group received the extract treatment for 12 weeks in conjunction with a high-fat diet sustained over a period of 24 weeks, n = 6), and N-FE (the preventive groups, specifically those adhering to a normal diet for 12 weeks followed by a high-fat diet for the subsequent 12 weeks, were concurrently subjected to extract treatment throughout the entire 12-week period, n = 6). The composition of the high-fat diet followed the Kartinah procedure (Kartinah et al., 2021). The 100 mg/kg body weight dose-response of the ethanol extract of *C. sativum* seeds was used for 12 weeks in this experiment (Mima et al., 2020). At the end of the 24th week, the rats were anesthetized using a ketamine 7.5 mg/kg body weight and xylazine 0.4 mg/kg body weight injection intraperitoneally. All experiments were conducted in accordance with the Laboratory Animal Research Institute Guidelines for the Care and Use of Laboratory Animals, National Research Council, and approved by the Ethics Committee of the Faculty of Medicine, Universitas Indonesia (Number: KET-662/UN2.F1/ETIK/PPM.00.02/2022) on June 7, 2022.

N	Acclimatation (normal diet)	Normal diet	Normal diet
F		High-fat diet	High-fat diet
NE		Normal diet	Normal diet + *C. sativum* extract
FE		High-fat diet	High-fat diet + *C. sativum* extract
N-FE		Normal diet	High-fat diet + *C. sativum* extract
Feeding time (weeks)	1	12	12

Figure 1. Experimental groups and experimental procedures.

Malondialdehyde (MDA) measurement

The thiobarbituric acid assay method was applied to the MDA measurement by spectrophotometry at a wavelength of 530 nm (Wills method) (Prijanti et al., 2018). The protein from tissue homogenates was previously precipitated with the addition of trichloroacetic acid. The results were expressed by nmol/mg tissue. The experiments were replicated twice.

Specific activity of catalase measurement

The activity of the catalase enzyme was determined by adding H_2O_2 1:4000 to the sample. The absorbance was read at 30 and 150 seconds by spectrophotometry at a 210 nm wavelength. The catalase enzyme activity was determined using the formula [1].

$$\frac{[\Delta \text{ sample-}\Delta \text{ blank/minute}]}{[\text{molarity of } H_2O_2 \times \text{volume of sample}]} \times \text{sample dilution} \quad [1]$$

The total protein of each sample was measured using the Warburg–Christian method. The specific activity of catalase was expressed by the catalase activity unit per mg total protein (Hardiany et al., 2022a). The experiments were replicated twice.

Glutathione (GSH) measurement

The Ellman method was used to estimate the GSH concentration in a sample. The assay was based on the oxidation reaction of GSH with DTNB (5,5′-dithiobis(2-nitrobenzoic acid)), which produced the TNB (5′-thio-2-nitrobenzoic acid) chromophore. The formation rate of TNB, measurable at 412 nm, was proportional to the GSH concentration in the sample. The GSH level was expressed as µg GSH per mg total protein (Prijanti et al., 2018; Hardiany et al., 2022a). The experiments were replicated twice.

Senescence-associated β-galactosidase (SA-β-Gal) assay

The SA-β-Gal assay was measured by the fluorometric method. A 10 mg tissue was homogenized in 200 µL of the cell lysis buffer, as in the kit instructions (96-Well Cellular Senescence Assay Kit (SA-β-Gal Activity, Fluorometric Format) [Cell Biolabs, Inc. CBA-231]). The lysate was transferred to a 96-well fluorescence plate and then added to a fresh assay buffer with equal volumes. The well was incubated at 37°C in the dark for 3 hours. The fluorescence measurement was 360 nm (excitation)/450 nm (emission). The measurement of total protein was performed using the Bradford method. The experiments were replicated twice.

The p21-level assay

The p21 level was assayed in tissue lysates using the Rat p21 Protein Elisa Kit according to the manual instructions (Rat p21 Protein Elisa Kit [MyBioSource 2602846]). The sample preparation used 10 mg of tissue homogenized in 600 µL of nuclei lysis solution (Promega). The wavelength of the photometric reading was set at 450 nm. The measurement of total protein was performed using the Bradford method. The experiments were replicated twice.

Statistical analysis

The determination of significant differences by analysis of variance (ANOVA) was performed and visualized with Graphpad. The results were presented as mean ± standard error of the mean (SEM). Tukey post hoc tests were used for multiple comparisons. A significant difference was established if $p<0.05$. A Pearson correlation analysis was conducted using the Statistical Package for the Social Sciences (SPSS) to examine the correlations among MDA concentration, glutathione level, and catalase activity.

RESULTS

Phytochemical screening of ethanolic *C. sativum* seed extract

The phytochemical compound screening of the *C. sativum* extract was evaluated by GC-MS. Based on the data (Table 1), several compounds were classified into the following groups: fatty acids (valeric acid, caproic acid, enanthic acid, caprylic acid, pelargonic acid, capric acid, lauric acid, pentadecylic acid, palmitelaidic acid, palmitic acid, oleic acid, petroselinic acid, vaccenic acid, stearic acid, and myristic acid), phenol (isoeugenol), and alcohol (1-monooleoylglycerol). The major components in the GC-MS chromatogram were palmitic acid (17.6079%) and vaccenic acid (37.0916%), as shown in the chromatogram (Fig. 2).

Oxidative stress and antioxidant level

MDA is an oxidative stress marker of lipid peroxidation. The MDA concentration increased in HFD feeding compared to the normal diet group (Fig. 3A), while the GSH level (Fig. 3B) and catalase-specific activity (Fig. 3C) decreased significantly. The MDA concentration negatively correlated with GSH level and catalase activity, as depicted in Fig. 4A-B. This implies that an elevation in MDA was countered by a decrease in GSH and catalase activity, substantiating a reduction in these antioxidants.

Table 1. GC-MS analysis of phytochemical compounds.

Compounds	IUPAC name	Molecular formula	Retention time	% Area
Valeric acid	Pentanoic acid	$C_5H_{10}O_2$	7.0575	1.6933
Caproic acid	Hexanoic acid	$C_6H_{12}O_2$	8.5824	1.6776
Enanthic acid	Heptanoic acid	$C_7H_{14}O_2$	10.0822	1.7325
Caprylic acid	Octanoic acid	$C_8H_{16}O_2$	11.5315	3.3952
Pelargonic acid	Nonanoic acid	$C_9H_{18}O_2$	12.9305	0.9702
Capric acid	Decanoic acid	$C_{10}H_{20}O_2$	14.2160	0.4051
Isoeugenol	2-methoxy-4-[E-prop-1-enyl]phenol	$C_{10}H_{12}O_2$	14.3672	2.0376
Lauric acid	Dodecanoic acid	$C_{12}H_{24}O_2$	16.5223	1.1424
Myristic acid	Tetradecanoic acid	$C_{14}H_{28}O_2$	18.6144	1.2521
Pentadecylic acid	Pentadecanoic acid	$C_{15}H_{30}O_2$	19.6101	0.1535
Palmitelaidic acid	(E)-hexadec-9-enoic acid	$C_{16}H_{30}O_2$	20.3537	0.3875
Palmitic acid	Hexadecanoic acid	$C_{16}H_{32}O_2$	20.5427	17.6079
Margaric acid	Heptadecanoic acid	$C_{17}H_{34}O_2$	21.4753	0.1807
Petroselinic acid	(Z)-octadec-6-enoic acid	$C_{18}H_{34}O_2$	21.7778	0.4043
Oleic acid	(Z)-octadec-9-enoic acid	$C_{18}H_{34}O_2$	21.9164	0.6804
Vaccenic acid	11-Octadecenoic acid, (Z)-	$C_{18}H_{34}O_2$	22.1307	37.0916
Stearic acid	Octadecanoic acid	$C_{18}H_{36}O_2$	22.3575	4.8257
Arachidic acid	Icosanoic acid	$C_{20}H_{40}O_2$	23.5296	0.4072
1-Monooleoylglycerol	2,3-dihydroxypropyl (Z)-octadec-9-enoate	$C_{21}H_{40}O_4$	26.3779	0.3764

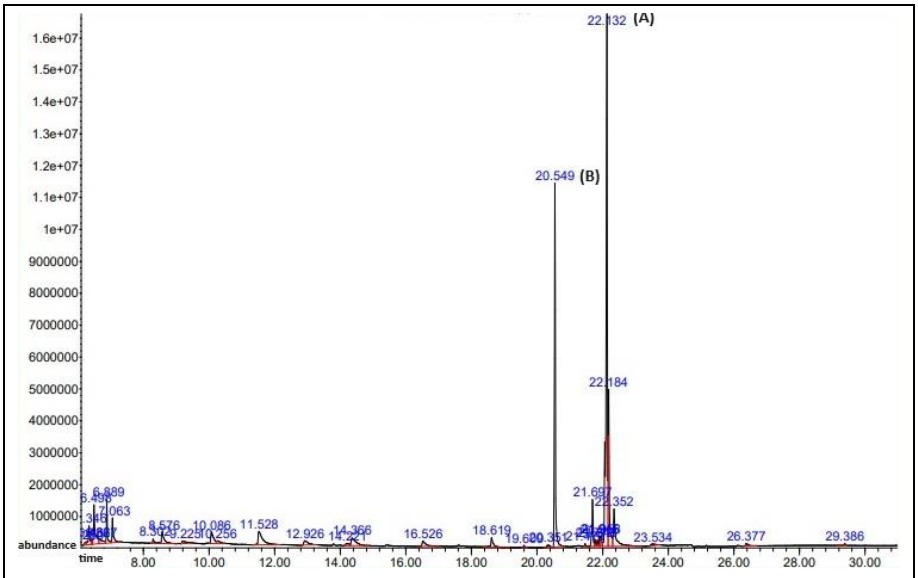

Figure 2. Chromatograms of the ethanol-based *C. sativum* extract.
The highest levels of components were vaccenic acid **(A)** and palmitic acid **(B)**.

This phenomenon is evident in the high-fat diet (HFD) groups, where an excess of fatty acid intake leads to elevated MDA production, followed by a decline in antioxidant enzyme levels. Consequently, we infer that the 24-week HFD feeding regimen induced oxidative stress.

A contrasting trend was observed with the extract treatment in both the normal and high-fat diets (HFD) conditions. The study revealed a significant reduction in MDA concentration due to the extract treatment in the normal diet group compared to both control groups. Similarly, a notable decrease in MDA concentration was evident with the extract treatment in the HFD group compared to the control HFD group, as illustrated in Fig. 3A.

Notably, the extract treatment demonstrated a significant difference in MDA reduction across all groups when compared to the control HFD group. This pattern was also observed in the preventive group (NFE), where MDA levels significantly decreased compared to the fat control group (F), accompanied by a concurrent increase in glutathione and catalase activity. Moreover, concerning glutathione (GSH) levels and catalase activity, a significant difference was observed in the normal diet group treated with ethanolic *C. sativum* seed extract compared to the control group, as well as in the preventive group compared to the fat control group (F).

Senescence marker

The statistical data indicate no significant differences in senescence markers among the groups, as shown in Fig. 5. However, a noticeable trend in the high-fat diet group (F) revealed an increase in SA-β-Gal activity compared to the normal diet group (N), which was not significant. In contrast, the ethanolic *C. sativum* seed extract reduced SA-β-Gal in the preventive group (NFE) and decreased the p21 levels in both the preventive (NFE) and obese rat (FE) groups compared to the fat control (F) group; however, it was not significant.

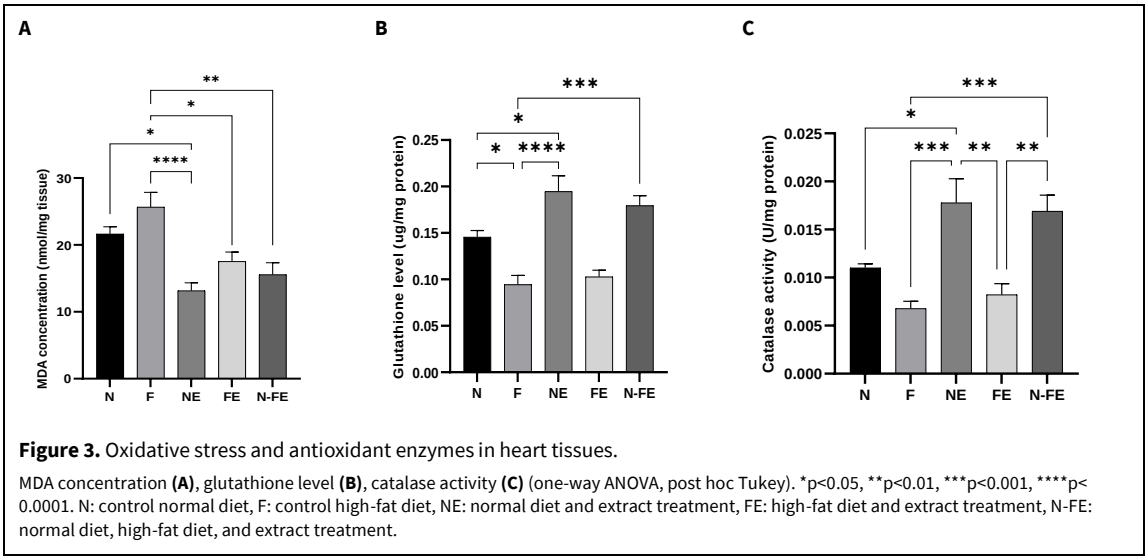

Figure 3. Oxidative stress and antioxidant enzymes in heart tissues.

MDA concentration **(A)**, glutathione level **(B)**, catalase activity **(C)** (one-way ANOVA, post hoc Tukey). *p<0.05, **p<0.01, ***p<0.001, ****p< 0.0001. N: control normal diet, F: control high-fat diet, NE: normal diet and extract treatment, FE: high-fat diet and extract treatment, N-FE: normal diet, high-fat diet, and extract treatment.

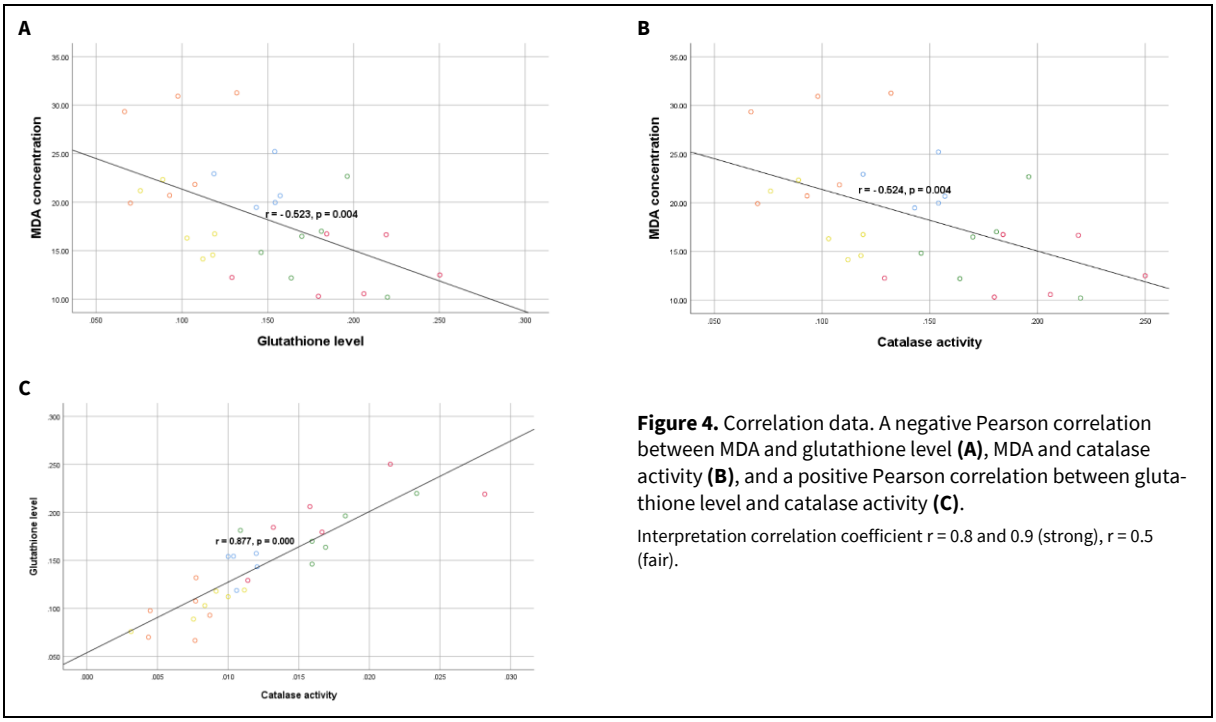

Figure 4. Correlation data. A negative Pearson correlation between MDA and glutathione level **(A)**, MDA and catalase activity **(B)**, and a positive Pearson correlation between glutathione level and catalase activity **(C)**.

Interpretation correlation coefficient r = 0.8 and 0.9 (strong), r = 0.5 (fair).

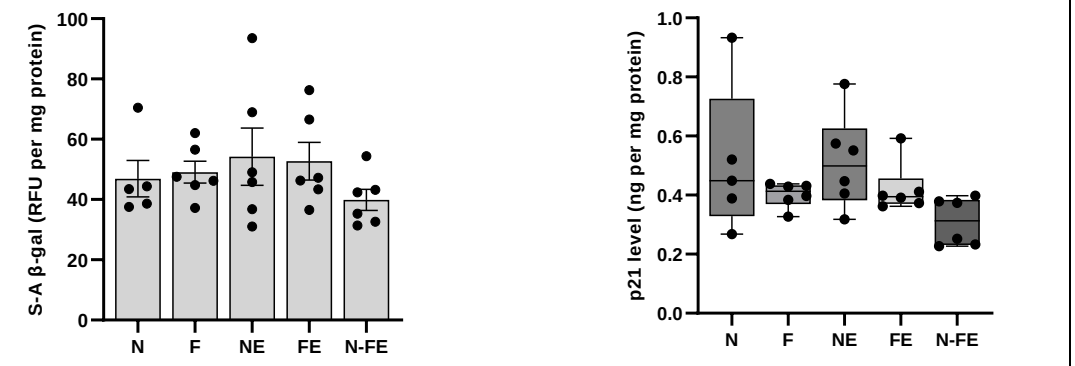

Figure 5. Senescence markers in heart tissue.

(A) Senescence-associated β-galactosidase activity (one-way ANOVA, data were mean ± SEM plotted, p = 0.4738). **(B)** p21 level (Kruskal-Wallis, data were median ± SEM plotted. p = 0.0625). N: control normal diet, F: control fat diet, NE: normal diet and extract treatment, FE: fat diet and extract treatment, N-FE: normal diet–fat diet and extract treatment.

DISCUSSION

Oxidative stress occurs when the production of ROS exceeds the antioxidant defense. Oxidative stress promotes lipid peroxidation, which is implicated in cell senescence. The structure and function of proteins, lipids, and nucleic acids are impaired by oxidizing ROS, leading to the dysfunction of those macromolecules, including the active sites of the enzymes, which were implicated in the accuration of substrate binding (Jiang et al., 2021). Carbonyl aldehyde groups of MDA can easily react with the end amino groups of proteins, forming intra- and intermolecular cross-links and resulting in biological effects, such as inactivating membrane transporters (Lankin et al., 2022).

This study demonstrated that a high-fat diet of saturated fatty acids induced oxidative stress, manifested as MDA elevation, catalase reduction, and a significant decrease in glutathione. Palmitic acid, a saturated fatty acid, is a potent inducer of ROS in cardiomyocytes (Wang et al., 2019). Fatty acids promote the elevation of oxidative stress by reducing catalase activity and sustaining the catalase protein level (Azevedo-Martins and Curi, 2008). In our study, the administration of a high-fat diet-induced obesity in rats, as demonstrated in our earlier investigation (Hardiany et al., 2022b). This obesity was subsequently accompanied by a reduction in glutathione (GSH) levels, aligning with the findings of Ozata et al. (2002). Antioxidant enzymes, such as catalase, superoxide dismutase, and glutathione, as the first line of defense against ROS, have a role in maintaining intracellular redox homeostasis, and a decrease in their activities contributes to oxidative damage to the tissues (Li et al., 2017).

The administration of *C. sativum* seed extract in our study demonstrated the capacity to reduce oxidative stress and elevate antioxidant levels. We propose that various compounds within the ethanolic *C. sativum* seed extract may be involved in activating the nuclear factor erythroid 2-related factor 2 (Nrf2), a key transcriptional master regulator that responds to oxidative stress (Ngo and Duennwald, 2022). Certain natural or synthetic electrophilic compounds target the binding of Kelch-like ECH-associated protein 1 (Keap1), eliciting a cellular response and fostering tolerance to endogenous stress (Abrescia et al., 2020).

The ethanolic *C. sativum* seed extract comprises monounsaturated fatty acids (such as vaccenic acid, oleic acid, and palmitelaidic acid) and saturated fatty acids (including palmitic acid, stearic acid and others), as determined by GC-MS examination. Notably, monounsaturated fatty acids, with vaccenic acid being a major component in our *C. sativum* seed extract, are likely responsible for activating Nrf2. This activation, in turn, contributes to an elevation in antioxidant levels and a reduction in MDA concentration. An unsaturated fatty acid structure's double band, susceptible to free radical attack, acts as an antioxidant to protect the lipid membrane (Ayala et al., 2014). Lipid peroxidation products activate Nrf2 for cellular redox homeostasis. In addition, a study reported that oleic acid potently increased the rates of complete fatty acid oxidation through activation of the sirtuin1–peroxisome proliferator-activated receptor-gamma coactivator-1alpha (SIRT1-PGC1α) mechanism (Lim et al., 2013). Complete oxidation results in the prevention of lipid peroxidation and MDA. We suggest that the oleic acid-containing extract decreases lipid peroxidation rates and is implicated in a decrease in MDA. Under low lipid peroxidation rates, the cells stimulate adaptive stress responses through the antioxidant defense system or signal pathway activation (Ayala et al., 2014).

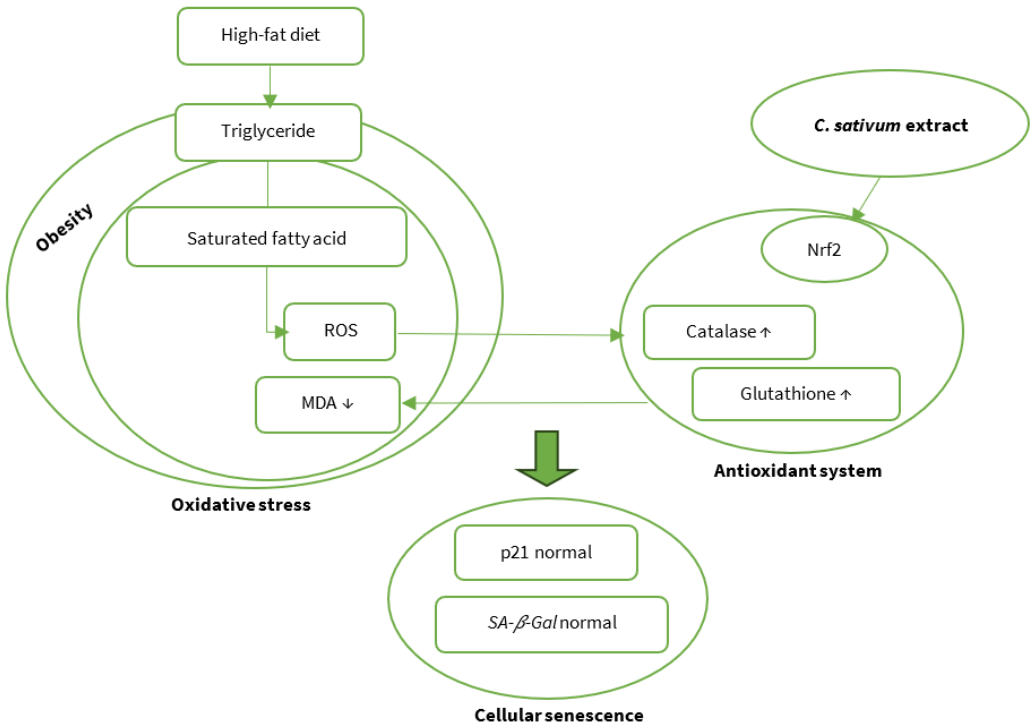

Figure 6. Proposed mechanism of action.

C. sativum extract stimulates Nrf2, which increases catalase activity and glutathione levels aimed at overcoming increased ROS, which is a product of lipid oxidation. MDA is reduced as a result of this mechanism. The process of cellular senescence is not impacted by this system.

Another extract-containing compound is isoeugenol, an isomeric of eugenol classified as terpenoid, and both are involved in reducing intracellular ROS and activating Nrf-2. The mechanism underlying the anti-oxidant activity of eugenol may involve cyclooxygenase-2 (COX-2) inhibition or the direct trapping of ROS molecules and enhanced stabilization and translocation nuclear of Nrf-2 (Ma et al., 2021). A study showed that isoeugenol activated Nrf2, displayed antioxidant and anti-inflammatory effects, and reduced the levels of Aβ peptides in *in vitro* and *in vivo* models of Alzheimer's disease (Silva et al., 2023).

Despite oxidative stress in the fat control group, no significant differences were observed in SA-β-Gal activity and p21 among the groups, indicating the absence of cellular senescence. This phenomenon may be attributed to the postmitotic nature of cardiomyocytes, preventing them from undergoing senescence since they are terminally differentiated. The fate of these cells is wholly contingent on their capacity to manage stress (Vicencio et al., 2008). Additionally, we propose that adaptive responses to oxidative stress play a role in maintaining SA-β-Gal activity and p21 levels (Fig. 6). p21 is crucial in initiating senescence in response to oxidative stress (Hamsanathan and Gurkar, 2022). A study also showed that p21 is integral to the antioxidant response, activating Nrf2 through Nrf2 protein stabilization (Chen et al., 2009). The interaction between p21 and Nrf2 results in Nrf2 stabilization, creating competition with Keap1 binding and preventing Nrf2 degradation (Ngo and Duennwald, 2022). Moreover, one study reported that Nrf2 directly binds to the p21 promoter, leading to the positive transcriptional regulation of p21 in response to low and moderate oxidative stress in cells (Jana et al., 2018).

CONCLUSION

This study proposed that ethanol-based *C. sativum* seed extract can potentially maintain heart health and reduce oxidative stress. However, its efficacy in inhibiting cellular senescence has not been conclusively demonstrated. The significant increase in antioxidant levels observed in normal rats receiving both standard and high-fat diets (preventive group) implies that *C. sativum* seed extract may serve as a preventive measure. Further research is needed to identify and elucidate the active compounds in *C. sativum* seed extract that exhibit potent antioxidant properties.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

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REFERENCES

Abrescia P, Treppiccione L, Rossi M, and Bergamo P (2020) Modulatory role of dietary polyunsaturated fatty acids in Nrf2-mediated redox homeostasis. Prog Lipid Res 80: 101066. https://doi.org/10.1016/j.plipres.2020.101066

Ayala A, Muñoz MF, Argüelles S (2014) Lipid peroxidation: production, metabolism, and signaling mechanisms of malondialdehyde and 4-hydroxy-2-nonenal. Oxid Med Cell Longev 2014: 360438. https://doi.org/10.1155/2014/360438

Azevedo-Martins AK, Curi R (2008) Fatty acids decrease catalase activity in human leukaemia cell lines. Cell Biochem Funct 26: 87–94. https://doi.org/10.1002/cbf.1404

Bhandari U, Kumar V, Khanna N, Panda BP (2011) The effect of high-fat diet-induced obesity on cardiovascular toxicity in Wistar albino rats. Hum Exp Toxicol 30: 1313-1321. https://doi.org/10.1177/0960327110389499

Birse RT, Choi J, Reardon K, Rodriguez J, Graham S, Diop S, Ocorr K, Bodmer R, Oldham S (2010) High-fat-diet-induced obesity and heart dysfunction are regulated by the TOR pathway in *Drosophila*. Cell Metab 12: 533–544. https://doi.org/10.1016/j.cmet.2010.09.014

Chen MS, Lee RT, Garbern JC (2022) Senescence mechanisms and targets in the heart. Cardiovasc Res 118: 1173–1187. https://doi.org/10.1093/cvr/cvab161

Chen W, Sun Z, Wang XJ, Jiang T, Huang Z, Fang D, Zhang DD (2009) Direct interaction between Nrf2 and p21(Cip1/WAF1) upregulates the Nrf2-mediated antioxidant response. Mol Cell 34: 663–673. https://doi.org/10.1016/j.molcel.2009.04.029

Davalli P, Mitic T, Caporali A, Lauriola A, D'Arca D (2016) ROS, cell senescence, and novel molecular mechanisms in aging and age-related diseases. Oxid Med Cell Longev 2016: 3565127. https://doi.org/10.1155/2016/3565127

Fang CX, Dong F, Thomas DP, Ma H, He L, Ren J (2008) Hypertrophic cardiomyopathy in high-fat diet-induced obesity: role of suppression of forkhead transcription factor and atrophy gene transcription. Am J Physiol Heart Circ Physiol 295: H1206-H1215. https://doi.org/10.1152/ajpheart.00319.2008

Hamsanathan S, Gurkar AU (2022). Lipids as regulators of cellular senescence. Front Physiol 13: 796850. https://doi.org/10.3389/fphys.2022.796850

Hardiany NS, Karman A, Calista A, Anindyanari BG, Rahardjo DE, Novira PR, Taufiq RR, Imtiyaz S, Antarianto RD (2022a) The effect of fasting on oxidative stress in the vital organs of New Zealand white rabbit. Rep Biochem Mol Biol 11: 190–199. https://doi.org/10.52547/rbmb.11.2.190

Hardiany NS, Lima FVId, Dewi S, Namirah I, Fadilah (2022b) The role of coriander seed extract (*Coriandrum sativum* L.) on cholesterol and blood plasma glucose of obese rats. [Indonesian]. J Biotek Medisiana Indonesia 2: 93-101.

Hsu HC, Chen CY, Lee BC, Chen MF (2016) High-fat diet induces cardiomyocyte apoptosis via the inhibition of autophagy. Eur J Nutr 55: 2245-2254. https://doi.org/10.1007/s00394-015-1034-7

Jana S, Patra K, Jana J, Mandal DP, Bhattacharjee S (2018) Nrf-2 transcriptionally activates P21Cip/WAF1 and promotes A549 cell survival against oxidative stress induced by H_2O_2. Chem Biol Interact 285: 59–68. https://doi.org/10.1016/j.cbi.2018.02.030

Jiang S, Liu H, Li C (2021) Dietary regulation of oxidative stress in chronic metabolic diseases. Foods 10: 1854. https://doi.org/10.3390/foods10081854

Kartinah NT, Komara N, Noviati ND, Dewi S, Yolanda S, Radhina A, Heriyanto H, Sianipar IR (2021) Potential of *Hibiscus sabdariffa* Linn. in managing FGF21 resistance in diet-induced-obesity rats via miR-34a regulation. Vet Med Sci 8: 309–317. https://doi.org/10.1002/vms3.653

Lankin VZ, Tikhaze AK, Melkumyants AM (2022) Malondialdehyde as an important key factor of molecular mechanisms of vascular wall damage under heart diseases development. Int J Mol Sci 24: 128. https://doi.org/10.3390/ijms24010128

Lawless C, Jurk D, Gillespie CS, Shanley D, Saretzki G, von Zglinicki T, Passos JF (2012) A stochastic step model of replicative senescence explains ROS production rate in ageing cell populations. PLoS One 7: e32117. https://doi.org/10.1371/journal.pone.0032117

Li W, Tang R, Ouyang S, Ma F, Liu Z, Wu J (2017) Folic acid prevents cardiac dysfunction and reduces myocardial fibrosis in a mouse model of high-fat diet-induced obesity. Nutr Metab (Lond) 14: 68. https://doi.org/10.1186/s12986-017-0224-0

Lim JH, Gerhart-Hines Z, Dominy JE, Lee Y, Kim S, Tabata M, Xiang YK, Puigserver P (2013) Oleic acid stimulates complete oxidation of fatty acids through protein kinase A-dependent activation of SIRT1-PGC1α complex. J Biol Chem 288: 7117–7126. https://doi.org/10.1074/jbc.M112.415729

Ma L, Liu J, Lin Q, Gu Y, Yu W (2021) Eugenol protects cells against oxidative stress via Nrf2. Exp Ther Med 21: 107. https://doi.org/10.3892/etm.2020.9539

Mahleyuddin NN, Moshawih S, Ming LC, Zulkifly HH, Kifli N, Loy MJ, Sarker MMR, Al-Worafi YM, Goh BH, Thuraisingam S, Goh HP (2021) *Coriandrum sativum* L.: A review on ethnopharmacology, phytochemistry, and cardiovascular benefits. Molecules 27: 209. https://doi.org/10.3390/molecules27010209

Mária J, Ingrid Z (2017) Effects of bioactive compounds on senescence and components of senescence associated secretory phenotypes *in vitro*. Food Funct 8: 2394–2418. https://doi.org/10.1039/c7fo00161d

Mima Y, Izumo N, Chen J-R, Yang S-C, Furukawa M, Watanabe Y (2020) Effects of *Coriandrum sativum* seed extract on aging-induced memory impairment in Samp8 mice. Nutrients 12: 455. https://doi.org/10.3390/nu12020455

Mohamed-Yassin MS, Baharudin N, Abdul-Razak S, Ramli AS, Lai NM (2021) Global prevalence of dyslipidaemia in adult populations: A systematic review protocol. BMJ Open 11: e049662. https://doi.org/10.1136/bmjopen-2021-049662

Ngo V, Duennwald ML (2022) Nrf2 and oxidative stress: a general overview of mechanisms and implications in human disease. Antioxidants (Basel) 11: 2345. https://doi.org/10.3390/antiox11122345

Ozata M, Mergen M, Oktenli C, Aydin A, Sanisoglu SY, Bolu E, Yilmaz MI, Sayal A, Isimer A, Ozdemir IC (2002) Increased oxidative stress and hypozincemia in male obesity. Clin Biochem 35: 627-631. https://doi.org/10.1016/S0009-9120(02)00363-6

Prijanti AR, Marissa N, Paramita R, Humaira S, Nabila EN, Wijaya AE, Fadila AN, Purwosunu Y (2018) Analysis of oxidative stress markers malondialdehyde, glutathione, nitric oxide, and prorenin level in preeclampsia placental tissues. Asian J Pharm Clin Res 11: 158–161. https://doi.org/10.22159/ajpcr.2018.v11i1.18330

Santos AL, Sinha S (2021) Obesity and aging: Molecular mechanisms and therapeutic approaches. Ageing Res Rev 67: 101268. https://doi.org/10.1016/j.arr.2021.101268

Shimizu I, Minamino T (2019) Cellular senescence in cardiac diseases. J Cardiol 74: 313-319. https://doi.org/10.1016/j.jjcc.2019.05.002

Silva A, Silva S, Macedo J, Moreira P, Baptista D, Bicker J, Fortuna A, Liberal J, Rodrigues B, Resende R, Santos AE, Pereira C, Cruz MT (2023) Targeting Alzheimer's disease hallmarks with the Nrf2 activator isoeugenol, Qeios https://doi.org/10.32388/LUXM7W

Tan BL, Norhaizan ME (2019) Effect of high-fat diets on oxidative stress, cellular inflammatory response and cognitive function. Nutrients 11: 2579. https://doi.org/10.3390/nu11112579

Vicencio JM, Galluzzi L, Tajeddine N, Ortiz C, Criollo A, Tasdemir E, Morselli E, Ben Younes A, Maiuri MC, Lavandero S, Kroemer G (2008) Senescence, apoptosis or autophagy? When a damaged cell must decide its path--a mini-review. Gerontology 54: 92–99. https://doi.org/10.1159/000129697

Wang Y, Wang XJ, Zhao LM, Pang ZD, She G, Song Z, Cheng X, Du XJ, Deng XL (2019) Oxidative stress induced by palmitic acid modulates K(Ca)2.3 channels in vascular endothelium. Exp Cell Res 383: 111552. https://doi.org/10.1016/j.yexcr.2019.111552

Wang Z, Li L, Zhao H, Peng S, Zuo Z (2015) Chronic high fat diet induces cardiac hypertrophy and fibrosis in mice. Metabolism 64: 917–925. https://doi.org/10.1016/j.metabol.2015.04.010

Xiang Q, Tian F, Xu J, Du X, Zhang S, Liu L (2022) New insight into dyslipidemia-induce cellular senescence in atherosclerosis. Biol Rev 97: 1844-1867. https://doi.org/10.1111/brv12866

Zhou Y, Li H, Xia N (2021) The interplay between adipose tissue and vasculature: Role of oxidative stress in obesity. Front Cardiovasc Med 8: 650214. https://doi.org/10.3389/fcvm.2021.650214

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Concepts or ideas					x			x
Design	x				x			x
Definition of intellectual content	x				x	x	x	x
Literature search	x	x	x	x	x	x	x	x
Experimental studies	x	x	x	x	x	x	x	x
Data acquisition	x	x	x	x	x	x	x	x
Data analysis	x	x	x	x				
Statistical analysis	x	x	x	x				
Manuscript preparation	x				x	x		x
Manuscript editing	x				x	x	x	x
Manuscript review	x	x	x	x	x	x	x	x

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