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Qualitative analysis of the antioxidant, carbohydrates, and lipids enzymes inhibitory effects of *Coriandrum sativum* seeds; a member of Palestinian flora

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Qualitative analysis of the antioxidant, carbohydrates, and lipids enzymes inhibitory effects of *Coriandrum sativum* seeds; a member of Palestinian flora

Abstract

The medicinal properties of spice plants have been recognized for centuries due to the essential oils obtained from these plants, which are considered therapeutic agents and have shown various therapeutic effects. *Coriandrum sativum* (*C. sativum*) seed oil was extracted using the organic solvent method and then examined for its antidiabetic (using the anti-amylase assay), antiobesity (using the anti-lipase assay), and antioxidant activities (using the DPPH inhibition assay) by in vitro methods. As observed, *C. sativum* seed oil showed a strong anti-amylase effect with an IC₅₀ equal to 79.43 ± 1.50 µg/ml compared to acarbose, which had an IC₅₀ equal to 28.1 ± 1.13 µg/ml. The strong anti-elastase effect of *C. sativum* oil was also reported with an IC₅₀ equal to 25.08 ± 1.23 mg/ml compared with the IC₅₀ value of oleanolic acid (the IC₅₀ of oleanolic acid is 25.09 ± 0.09 µg/ml). In addition, potent antioxidant activity with an IC₅₀ equal to 17.78 ± 1.62 µg/ml was compared to the Trolox, which had an IC₅₀ equal to 2.7 ± 0.07 µg/ml. On the other hand, moderate to weak effects as antiobesity agents were reported with an IC₅₀ equal to 91.20 ± 1.27 µg/ml compared to orlistat, which had an IC₅₀ equal to 12.09 ± 0.09 µg/ml. This study confirms the traditional use of the seeds of *C. sativum* as a natural remedy for diabetes, oxidative stress conditions, and some skin disorders. Further in vivo investigation must be conducted in the future.

Keywords

Coriandrum sativum; anti-diabetic; anti-obesity; anti-elastase; antioxidant

Cover Page Footnote

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Qualitative analysis of the antioxidant, carbohydrates, and lipids enzymes inhibitory effects of *Coriandrum sativum* seeds; a member of Palestinian flora

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ABSTRACT

The medicinal properties of spice plants have been recognized for centuries due to the essential oils obtained from these plants, which are considered therapeutic agents and have shown various therapeutic effects. *Coriandrum sativum* (*C. sativum*) seed oil was extracted using the organic solvent method and then examined for its antidiabetic (using the anti-amylase assay), antiobesity (using the anti-lipase assay), and antioxidant activities (using the DPPH inhibition assay) by *in vitro* methods. As observed, *C. sativum* seed oil showed a strong anti-amylase effect with an IC₅₀ equal to 79.43 ±1.50 µg/ml compared to acarbose, which had an IC₅₀ equal to 28.1 ±1.13 µg/ml. The strong anti-elastase effect of *C. sativum* oil was also reported with an IC₅₀ equal to 25.08 ±1.23 mg/ml compared with the IC₅₀ value of oleanolic acid (the IC₅₀ of oleanolic acid is 25.09 ±0.09 µg/ml). In addition, potent antioxidant activity with an IC₅₀ equal to 17.78 ±1.62 µg/ml was compared to the Trolox, which had an IC₅₀ equal to 2.7 ±0.07 µg/ml. On the other hand, moderate to weak effects as antiobesity agents were reported with an IC₅₀ equal to 91.20 ±1.27 µg/ml compared to orlistat, which had an IC₅₀ equal to 12.09 ±0.09 µg/ml. This study confirms the traditional use of the seeds of *C. sativum* as a natural remedy for diabetes, oxidative stress conditions, and some skin disorders. Further *in vivo* investigation must be conducted in the future.

Keywords: *Coriandrum sativum*; anti-diabetic; anti-obesity; anti-elastase; antioxidant.

INTRODUCTION

The popularity of alternative medicine, such as herbal medicine, has grown significantly in both developed and less developed countries because the alternative therapies are "natural" they are significantly safer than prescription medicine (1, 2). Herbal extracts are rich in phytochemicals and bioactive ingredients that can work together through different mechanisms to give the estimated therapeutic effect (3). Additionally, it should be noted that the majority of pharmaceuticals originated in and developed from medicinal plants (4, 5). Almost 70% of current medicinal products in India are sourced from natural sources (6). In order to determine or enhance the therapeutic characteristics of the isolated molecule so that it may be used pharmacologically in both industrial and clinical contexts, researchers are

examining a broad range of herbs. In particular, they want to utilize this chemical in clinical and industrial contexts (7).

In cells, oxidative stress can create peroxides and free radicals, damaging anything from DNA to proteins and lipids (8). It is also believed that this stress can boost many diseases and conditions like autism, Alzheimer's disease, Asperger syndrome, atherosclerosis, cancer, attention deficit hyperactivity disorder, Lafora disease, Parkinson's disease, myocardial infarction, heart failure, Sickle Cell disease, fragile X syndrome, infectious conditions, vitiligo, depression, and chronic fatigue syndrome (9).

Diabetes mellitus (DM) is the most prevalent non-infectious disease globally. DM is a metabolic condition. Diabetes is caused by a lack of insulin synthesis, insulin action, or

both. In turn, insulin insufficiency causes persistent hyperglycemia and abnormalities in carbohydrate, lipid, and protein metabolism (10). Studies showed promising results in treating diabetes mellitus using inhibition of alpha-amylase (11). In general, obesity is thought to be a problem with lipid metabolism, and the enzymes involved in this process could be precisely targeted in the development of antiobesity drugs. As dietary triglycerides are the principal source of excess calories, new advancements in the treatment of obesity include the inhibition of pancreatic lipase to reduce dietary triglyceride absorption (PL). Natural products offer a vast reservoir of PL inhibitors that may be turned into therapeutic drugs (12, 13).

However, scientific progress that has unlocked the molecular dynamics of lipid metabolism has led to a greater understanding of the developing activities of lipid hydrolyzing enzymes such as pancreatic lipase. Delaying fatty acid absorption into the systemic circulation and adipocytes and slowing triglyceride breakdown are expected outcomes of suppressing pancreatic lipase (14, 15). In clinical settings, the only conventional pancreatic lipase enzyme inhibitor accessible is orlistat; hence, identifying safe therapeutic alternatives derived from plants that inhibit pancreatic lipase has been recognized as a significant achievement. Therefore, plants showing considerable promise in treating obesity are currently being examined for their capacity to suppress pancreatic lipase (15).

Coriandrum sativum (*C. Sativum*), also known as cilantro or Chinese parsley and in Arabic called Kuzbarah (16), is a plant that belongs to the *Umbelliferae* family (17). The plant's leaves are decomposed leaves that are

divided irregularly. It is an annual herb cultivated in dry weather, usually in March. It grows to a height of 1 to 3 feet (30–90 cm), is slender, and branched in shape. The best harvest time is in August, when the fruit is ripe and falling. This plant has an unpleasant, distinctive aroma and a spicy flavor (18).

The seed oil contains about 1% volatile oil (linalool or coriander) (19). Components such as borneol, camphor, p-cymene, limonene, geraniol, and alpha-pinenes are also present (20). The raw seed oil also contains terpinene and geranyl acetate (21). It can relieve flatulence and spasms, reduce excessive sweating, and increase appetite (22). It is also a stimulant, flavoring agent, tonic, and diuretic. It can be beneficial for rheumatism and painful joints (23).

Adding coriander seeds and leaves into the food will enhance the antioxidant content and thus probably prevent oxidative food deterioration (24). In addition, food shelf-life can be prolonged by adding antioxidants, and coriander is rich in these antioxidants, so seasoning food with this spice will protect it from spoilage (17). This project aimed to investigate the antioxidant, carbohydrates, and lipids enzymes inhibitory effects of *C. sativum* seeds oil.

MATERIALS AND METHODS

Materials

Trolox ((s)-(-)-6 hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) was obtained from (Sigma Aldrich, Denmark), and (DPPH) 2,2-Diphenyl-1-picrylhydrazyl was obtained from (Sigma Aldrich, Germany) (Sigma Aldrich, Germany). The solvents used for the extraction of plant material were of HPLC grade. Sigma-Aldrich (USA) provided

acarbose, orlistat, starch, PNPP (p-nitrophenyl palmitate), DPPH (2,2-diphenyl-1-picrylhydrazyl), and porcine pancreatic lipase, while MP Biochemicals (Illkirch, France) provided porcine pancreatic -amylase. Only analytical-grade substances were utilized in the tests. The *C. sativum* seeds used in this experiment originated from the Al-Saffarini Farm in Palestine. The plant seeds were examined in the Pharmacy Department of An-Najah National University, where they were cataloged as specimen Pharm-PCT-2777.

Extraction of the fixed oil

After measuring out 100 g of the powdered seeds, we dissolved them in a mixture of 200 ml of n-hexane, 400 ml of 50% ethanol, and 100 ml of triple-distilled water to extract the active ingredients. Then, using a 200 rpm shaker, the mixture was agitated for 72 hours at room temperature. The solution was then filtered using a suction flask and a Buchner funnel. The plant oil, which is in the organic phase, was then separated from the watery phase using a separatory funnel (lower one). After collecting the organic phase, it was dried in a rotary evaporator at 40 °C for one hour to remove the solvents, and the resulting substance was kept at room temperature until use. The leftover material was extracted once more with the addition of 50 ml of hexane and 125 ml of 50% ethanol to triple-distilled water. (25).

***α*-Amylase inhibitory assay**

A standard modification procedure was performed to evaluate the oil's *α*-amylase inhibitory action (26, 27). A few milliliters of 10% DMSO were used to dissolve the extraction, diluted in buffer (0.02 M Na₂HPO₄/NaH₂PO₄, 0.006 M NaCl, pH 6.9)

to produce a stock solution with a concentration of 100 µg/ml. Using 10% DMSO as the dilution solvent, the following dilutions were created: 10, 50, 70, 100, and 500 µg/ml. Then, 0.2 ml of porcine pancreatic -amylase enzyme (Type VI-B) solution (2 units/ml) was combined with 0.2 ml of coriander oil fraction and incubated for 10 minutes at 30 °C. Then, 0.2 ml of newly made 1% aqueous starch solution was added to each test tube and incubated for three minutes. 0.2 ml of DNSA reagent diluted with 5 ml of DW was added to halt the reaction. This combination should next be boiled at 90 °C for 10 minutes in a water bath and then cooled to ambient temperature. The finished mixture's absorbance was evaluated at 540 nm. In place of the oil, 0.2 ml of the final buffer was added to the preceding procedures to create the blank. In the same manner as acarbose, the standard reference was likewise prepared with acarbose. This equation (1) was utilized to evaluate the inhibitory activity of *α*-amylase.:

$$\alpha - \text{amylase inhibitory \%} = \frac{(A_B - A_T)}{A_B} \times 100\% \quad \text{Equation (1)}$$

A_B: Recorded absorbance of the blank solution

A_T: Recorded absorbance of the tested sample solution

Elastase inhibitory activity assay of C. sativum oil

The inhibitory effect of *C. sativum* oil on the skin-aging enzyme porcine pancreatic elastase (PPE) (Type I) was determined by spectrophotometric analysis (28). SANA was considered as the substrate. At a wavelength of 410 nm, the absorbance was measured to obtain the release of p-nitroaniline for 15 min at 25 °C. To carry out the reaction, a buffer of

2 mM Tris-HCl (pH 8.0) and elastase enzyme in a 10% DMSO solution with a concentration of 10 µg/ml, a five mM SANA, and a stock solution of 1000 µg/ml were prepared. A stock solution of 100 ml was prepared by dissolving 100 mg of coriander oil in 10% DMSO. After that, the following dilutions were prepared (10, 20, 30, 40, 50, 100, and 200 µg/ml). The mixture was then incubated for 10 minutes at 25 °C. It comprised 1 ml of each dilution and 5 ml of Tris-HCl buffer. After adding 0.5 ml of substrate SANA to each test tube, they were re-incubated at 25 °C for 15 minutes. Except for the coriander oil, the blank has all the elements from the preceding combination. Oleonic acid served as a positive control. To calculate the percentage of inhibition of the oil, the following equation (2) is used:

$$\text{Inhibition (\%)} = \frac{(A-B)}{A} \times 100\% \quad \text{Equation (2)}$$

A: Enzyme activity without an inhibitor

B: Enzyme activity in the presence of an inhibitor

Porcine pancreatic lipase enzyme inhibitory effect of C. sativum oil

First, a plant solution with a 1 mg/ml concentration was made by dissolving 100 mg of coriander oil in 100 ml of 10% DMSO. The sample was diluted to get the following concentrations: 50, 100, 200, 300, and 400 µg/ml. In addition, a freshly manufactured lipase enzyme (Type II) stock solution was created by dissolving 25 mg of lipase powder in 25 ml of 10% DMSO to produce a 1 mg/ml solution. Meanwhile, a stock solution of p-nitrophenyl butyrate (PNPB) was prepared by combining 20.9 mg of PNPB with 2 ml of acetonitrile. Then, 0.2 ml of the plant solution was serially diluted, followed by adding 0.1 ml of lipase stock solution and enough Tris-HCl solution

to produce 1 ml in total volume. The resulting solutions were then incubated for 15 minutes in a water bath maintained at 37 °C. After adding 0.1 ml of the PNPB solution, the mixture must be incubated at the same temperature for 30 minutes (27, 29). The following equation (3) was later used to calculate the absolute inhibitory activity.

$$\text{Inhibition (\%)} = \frac{(B-S)}{B} \times 100\% \quad \text{Equation (3)}$$

Where S refers to the sample absorbance and B to the blank absorbance.

Antioxidant activity of C. sativum oil

In the beginning, a methanolic stock solution was made for both coriander oil and Trolox to make a concentration of 1000 µg/ml for each one. Trolox was considered the standard reference as it has a strong antioxidant effect. Then, the stock solution underwent serial dilutions to reach the following concentrations (2, 3, 5, 7, 10, 20, 30, 50, 80, and 100 µg/ml). After that, 1 ml of each of the previous diluent solutions was placed in a test tube, followed by the addition of 1 ml of freshly prepared DPPH methanolic solution, which has a concentration of 0.002 g/ml. A DPPH methanolic solution is considered the most commonly used free radical. Then 1 ml of methanol was measured and added. To formulate the blank solution, in a 1:1 ratio, DPPH and methanol were blended. The previous solutions were then incubated for thirty minutes at room temperature (25 °C) in a dark condition. A spectrophotometer was used to evaluate the optical densities of these solutions at a wavelength of 517 nm. To figure out how much DPPH was blocked by previous mixtures (blank and plant solutions) (27, 30). Equation (4) below was used:

$$\text{DPPH inhibition\%} = \frac{(AB - AS)}{AB} \times 100\% \text{ Equation (4)}$$

AB: Blank solution absorbance

AS: Sample solution absorbance.

Statistical analysis

The findings were expressed as the mean \pm standard deviation (SD). The IC_{50} (50% Inhibition concentration) was calculated using BioDataFit edition 1.02 (30).

RESULTS

α -Amylase inhibitory results

C. sativum oil was tested for its ability to inhibit α -amylase and was compared to the positive control, acarbose. When compared with acarbose, as shown in (Figure 1), the oil showed inhibitory activity of α -amylase. The IC_{50} of *C. sativum* oil (IC_{50} 79.43 \pm 1.50 μ g/ml) was slightly close to the IC_{50} of acarbose (IC_{50} value of acarbose is 28.1 \pm 1.13 μ g/ml).

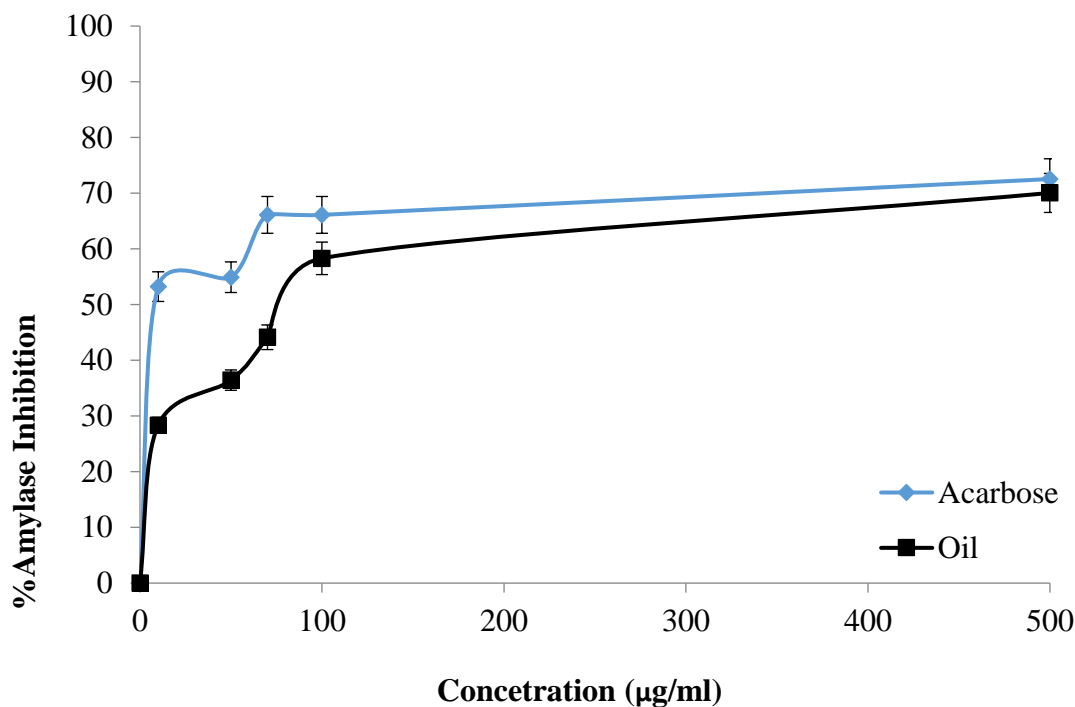


Figure (1): α -Amylase inhibitory activity of *C. sativum* oil and acarbose.

Elastase inhibitory activity results

A Spectrophotometer was used to examine the effect of *C. sativum* oil on the inhibition of PPE (Porcine Pancreatic Elastase). (Fig-

ure 2) showed that *C. sativum* oil's anti-elastase effect compared with oleanolic acid, as it had an IC_{50} value (IC_{50} of *C. sativum* oil is 25.08 \pm 1.23 mg/ml) approximately similar to the IC_{50} value of oleanolic acid (IC_{50} of oleanolic acid is 25.09 \pm 0.09 mg/ml).

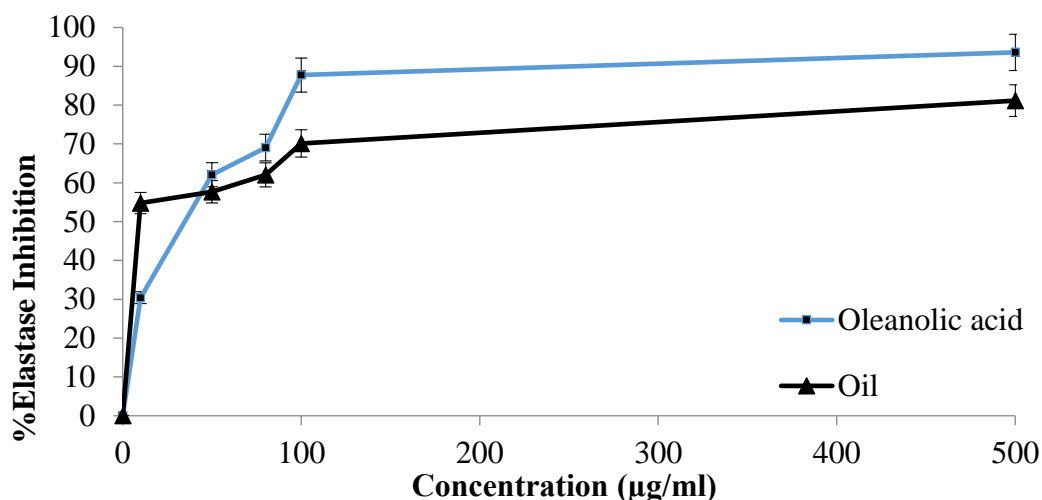


Figure (2): The percentages of elastase inhibitory activity for *C. sativum* and oleanolic acid.

Porcine pancreatic lipase enzyme inhibitory

The antilipase effect of *C. sativum* oil was obtained and compared with the inhibitory effect of orlistat. (Figure 3) showed the antilipase impact of the oil compared with the

orlistat; the IC_{50} value of *C. sativum* oil was around $91.20 \pm 1.27 \mu\text{g/ml}$, while for orlistat, it was $12.09 \pm 0.09 \mu\text{g/ml}$. As a result, it inhibits the porcine pancreatic lipase enzyme in a weak-to-moderate manner.

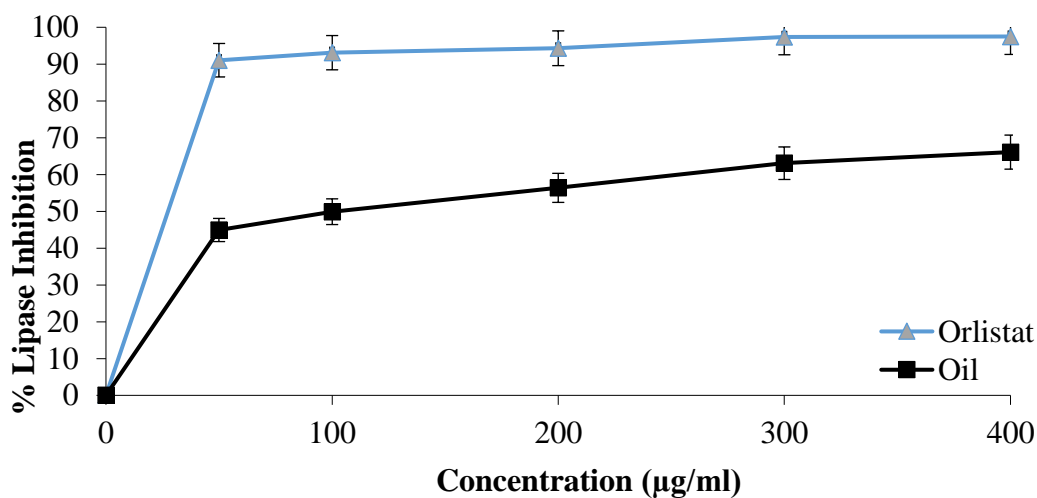


Figure (3): Porcine pancreatic lipase inhibitory activity of *C. sativum* oil and orlistat.

Antioxidant results

The antioxidant effect of *C. sativum* oil was obtained using the DPPH method. (Figure 4) showed the oil antioxidant activity in compared with Trolox (reference standard), in

which its IC₅₀ value (IC₅₀ of *C. sativum* oil, 17.78 ± 1.62 µg/ml) is very close to the IC₅₀ of Trolox (IC₅₀ of standard Trolox, 2.7 ± 0.07 µg/ml).

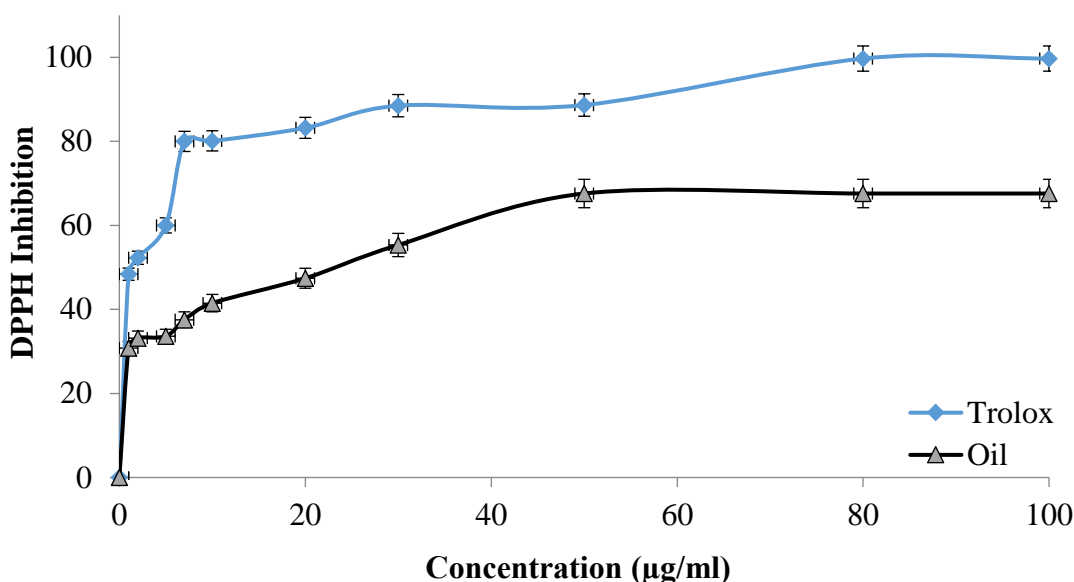


Figure (4): The percentages of DPPH inhibitory activity for *C. sativum* oil and Trolox.

DISCUSSION

Many studies were done to make sure that *C. sativum* was growing properly. For example, an aqueous extract of these seeds was said to be able to stop some bacterial strains from developing. Extracts made from the plant's leaves, seeds, and stems were found to have many antibacterial properties. The best results were found against *Klebsiella pneumoniae* and *Proteus mirabilis* (31).

It is now well known that oxidative stress contributes to the development, maintenance, and progression of diabetes and its complica-

tions (32). In most cases, diabetes is accompanied by an increase in free radical generation (33-35). *C. sativum* has much hypoglycemic power in high doses, and It can work efficiently in combination with oral hypoglycemic medications in type-2 diabetic individuals whose diabetes was not well managed by oral hypoglycemic medications alone (36, 37). *C. sativum* seed oil has a potent anti-amylase effect, with an IC₅₀ of 79.43 ± 1.50 mg/ml compared to the control compound, acarbose, which has an IC₅₀ of 28.1 ± 1.13 mg/ml. This confirms that this plant is suitable for treating diabetes.

Additional studies demonstrated that the high total phenolic content of *C. sativum* made it an effective antioxidant for scavenging 2,2-diphenyl-1-picrylhydrazyl (DPPH) radicals, inhibiting lipoxygenase and phospholipid peroxidation, chelating iron, neutralizing hydroxyl radicals, disrupting superoxide radicals, lowering glutathione levels, and preventing lipid peroxidation (38). Coriander was shown to have antioxidant properties, suggesting it may be used to prevent harmful oxidation reactions (39). Our research confirmed these previous observations as we found a potent antioxidant activity with an IC_{50} equal to 17.78 ± 1.62 $\mu\text{g/ml}$ compared to the Trolox (the control compound), which has an IC_{50} equal to 2.7 ± 0.07 $\mu\text{g/ml}$ according to the listed results in this research.

When compared to a combination of oils that also includes coriander oil, the anti-hypercholesterolemic effect of pure coriander seed oil appears to be more potent (40). As well as reported in our study, in the lipase inhibition assay, moderate to weak effects of antiobesity agents were reported with an IC_{50} equal to 91.20 ± 1.27 $\mu\text{g/ml}$ compared to orlistat (the control compound), which has an IC_{50} equal to 12.09 ± 0.09 $\mu\text{g/ml}$.

Studies have shown that the primary chemical contents of coriander seeds, including essential oils and monoterpenoids like linalool, have dermatological benefits, including anti-inflammatory action in erythema generated by ultraviolet (UV) radiation and antioxidant effects on the skin (41). Additionally, this oil has a high concentration of beneficial fatty acids, including palmitic acid, linoleic acid, and oleic acid, which all work to reduce inflammation, enhance wound healing, and keep skin moisturized (42, 43). This research

has been updated with new results for *C. sativum* seed oil on elastase enzyme inhibition power to make this plant a good choice for skin disorders natural remedies as we observed a strong anti-elastase effect of *C. sativum* oil with an IC_{50} equal to 25.08 ± 1.23 $\mu\text{g/ml}$ compared with the IC_{50} value of oleanolic acid (IC_{50} of oleanolic acid is 25.09 ± 0.09 $\mu\text{g/ml}$).

This investigation is limited to the isolation, chemical structure elucidation, and structure-activity relationship (SAR) determination of the most therapeutically active molecules from *C. sativum* oil. Nevertheless, these experiments will be our future objective.

CONCLUSION

Further investigations must be conducted to establish new natural remedies from this plant seed oil that may be used in manufacturing advanced medication that may be effective in curing different health problems, mainly as recorded in this research, as this plant seed oil showed powerful antioxidant and anti-elastase effects.

Consent for publication

The authors give the Publisher the Author's permission to publish the work.

Data Availability

All data generated for this study are included in the article.

Author Contributions

Ahmad Eid: Conceptualization, writing-original draft, data curation, formal analysis, investigation, methodology, project administration, resources, software, supervision, validation, visualization, and writing review & editing. **Nidal Jaradat:** Writing-original draft, data formal analysis, investigation,

methodology. **Linda Issa:** Writing-original draft, data curation, formal analysis. **Essa Khraiwesh:** Writing-original draft, data curation. **Isra' Yaish:** Writing-original draft, data curation. **Zeinah Amleh:** Writing-original draft, data curation.

Competing Interest

The authors state that they have no known competing financial interests or personal relationships that might have influenced the research presented in this study.

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