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Influence of germicidal ultraviolet radiation UV-C on the quality of *Apiaceae* spices seeds

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Abstract

Background Spices are susceptible to surface microbial contamination. Countries' ban on ethylene oxide fumigation due to possible residual toxicity encouraged the usage of irradiation. Surface sterilization with low doses of ultraviolet radiation has been extensively researched as a safe, eco-friendly, and fast route. This study examines the quality of *Apiaceae* spices for consumption, including anise, fennel, caraway, and cumin, in response to germicidal ultraviolet radiation using a developed sterilization unit.

Methods The influence of UV-C (254 nm, 10.5 mW/cm^2) on the fungal and microbial count, germination percentage, respiration rate, phenolic content, essential oil, and the activity of antioxidant enzymes was investigated at exposure durations of 0–45 min in increments of 5 min. The treated seeds were packed in polyethylene bags in a naturally aerated storage room for 30 days before the inspection.

Results The obtained data showed that UV-C stimulated seeds germination and increased respiration rate for all studied types. The 25 min of exposure exhibited the highest significant values compared to the control, considered a good indicator of seed vigor. In addition, UV-C exposure between 20 and 35 min promoted the accumulation of phenolic compounds and increased the oil content as a defense mechanism against radiation. Conversely, higher exposure to UV-C led to a significant reduction in phenolic and oil contents. Furthermore, the exposure to UV-C radiation enhanced the activity of antioxidant enzymes in terms of peroxidase and catalase, which progressively increased with increasing exposure durations, reached their peak at 25–30 min, and subsequently declined with extended exposure time was extended. In a similar pattern, exposure to UV-C radiation increased polyphenol oxidase activity to its highest level at 25 min, owing to the development of antioxidant protective mechanisms against oxidative stress.

Conclusion UV-C irradiation in the range of 25–30 min is the most appropriate pretreatment to maintain the vitality of the examined seeds.

Keywords Spices, Ultraviolet radiation, Antioxidant activity, essential oil

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Graphical Abstract

Introduction

Spices and herbs are commonly consumed in processed foods worldwide for their powerful flavor, nutritional values, and medicinal characteristics [1]. Spices and herbs are non-perishable due to their low moisture content; nevertheless, microbial species rapidly grow when mixed with water-rich foods, causing the food to degrade quickly. Hence, spices are easily microbially contaminated by dust, wastewater, and animal/human excreta during numerous operations of cultivation, harvesting, drying, transportation, and storage [2–4]. The most common causes of foodborne infection in medicinal plants are toxic molds [5] and pathogenic bacteria [6]. Furthermore, many investigations have reported the presence of aflatoxin in a variety of herbs and spices, including black peppers, paprika, chili, ginger, anise, fennel, and cumin, that have been contaminated with diverse toxigenic species, such as *Aspergillus*, *Penicillium*, and *Rhizopus* [5, 7, 8].

Commercially, fumigation with ethylene oxide is used to lower microbial load infection in spices after extensive studies indicated that it has both antibacterial and insecticidal effects. [9]. Nevertheless, the European Union has outlawed fumigation due to the toxicity of the breakdown products and the residues' carcinogenic effect. In addition, heated steam serves as an antimicrobial treatment; however, high-temperature steam causes discoloration, flavor loss, volatile oil loss, and quality decline [10, 11].

The restriction on ethylene oxide in the European Union and other nations encouraged the use of irradiation. Currently, microwave, gamma radiation, X-rays, and electron beam applications are being used to suppress contamination and prolong the shelf-life of medicinal plants [6, 12]. However, ionizing radiation is restricted due to the high cost, complicated processing, market acceptance, and changes in spices taste [12, 13]. In general, heating processes cause significant damage owing to non-enzymatic browning, which leads to the Maillard reaction and the loss of color, nutritional value, and aroma [14].

On the other hand, UV-C sterilization inactivates bacteria and viruses efficiently [15]; due to mutations in the microorganisms' DNA nucleic acid, resulting in a reduction in the microbial population curve [16], with no concerns about residuals or disinfection by-products of UV-C [17]. According to Hidaka and Kubota [18] and Manzocco et al. [19], UV-C disinfection affects only the surface, which is sufficient to maintain spices hygiene, because most bacteria that contaminate spices reside on the surface. At the same time, the inner sections usually are contamination free [20]. The effect of germicidal ultraviolet light on regulating decay and physiological disorders without changing quality has been extensively researched on vegetables, fruits, milk, and juices [21–25].

Oxidative stress is caused by an imbalance in the oxidant/antioxidant system, which is revealed by a constant

increase in reactive oxygen species (ROS). Ultraviolet radiation energy can act as an abiotic stress elicitor in plants, promoting the accumulation and biosynthesis of bioactive components that directly minimize oxidative stress by scavenging free radicals.

According to Castillejo et al. [26], UV abiotic stresses have positive effects at low doses, including the synthesis of secondary metabolites such as carotenoids, phenolics, and flavonoids, as well as the production of antioxidant enzymes that protect plants against oxidative damage, such as the membrane lipid peroxidation [27, 28]. As UV exposure stimulates the generation of reactive oxygen species (ROS), it has been used to study the behavior of the antioxidant defense system of the plant. According to Costa et al. [29], UV-B radiation stimulated the antioxidant defense system in sunflower cotyledons. Moreover, UV-B radiation strengthened the antioxidant system in barley seedlings by increasing the activity of antioxidant enzymes and causing the formation of phenolic acids [28]. The plant's antioxidant defense mechanism is influenced by the type of stressor, as well as the severity and duration of its action [30]. To the best of our knowledge, the effectiveness of decontaminated short-wave UV radiation (UV-C) on the quality of spices seeds for consumption has not been adequately discussed. The goal of this study is to determine the optimal UV-C dosage that has a positive influence on the quality of *Apiaceae* spices for consumption after 30 days of storage. Anise (*Pimpinella anisum* L.), fennel (*Foeniculum vulgare* Mill), caraway (*Carum carvi* L.), and cumin (*Cuminum cyminum* L.) were examined for fungal and microbial count, germination percentage, respiration rate, phenolic content, essential oil, as well as the defense mechanism through the enzyme's activity.

Materials and methods

UV-C sterilization unit

UV-C unit was designed and developed according to Hidaka and Kubota [18] with some modifications to conduct the studied species' sterilization process. It was carried on a custom-designed iron chassis with dimensions of (125 × 65 × 71 cm^3; length × width × height). A sterilization tunnel measures (100 × 45 × 34 cm^3; length × width × height) was equipped with a transparent belt conveyor covered with trophic stainless-steel wire 30 cm wide and controlled by a 1.5 kW electric motor. Four UV-C lamps (UVC T8 30 W G13, LEDVANCE—China; 12.6 W) were assembled on the tunnel, two of which were hung at 20 cm from the belt [31], and the others were assembled under the belt at a distance of 13.5 cm. The distances between the belt and the lamps were assessed based on laboratory experiments to ensure a uniform radiation intensity across the entire surface of the seeds, using a digital radiometer (UV-37SD Data Logger Ultraviolet Intensity Meter, CUSTOM, Japan).

A stainless-steel hopper with a capacity of 5 kg and a spiral grooved wheel metering device at the bottom was positioned over the outer part of the feeding belt to feed the seeds uniformly in the required amount [32]. Meanwhile, a stainless-steel discharge hopper of similar capacity was installed under the moving belt at the end of the unit chassis to receive the treated seeds. Tunnel exits were insulated with curtain sheets to block any leak of UV radiation out of the tunnel.

Sample selection and UV-C treatment

Anise (*Pimpinella anisum* L.), fennel (*Foeniculum vulgare*), caraway (*Carum carvi* L.), and cumin (*Cuminum cyminum* L.) seeds were purchased from a local company located in the Cairo government. The UV-C unit was operated for the sterilization process, and the belt was set to the required speed to feed a thin layer of seeds over the belt surface. The seeds were placed in the feeding hopper, and the feeding hopper door was opened, allowing the seeds to descend onto the belt under the ultraviolet area. Each type of spices under investigation was irradiated with UV-light (254 nm, 10.5 mW/cm^2) for different durations of 5, 10, 15, 20, 25, 30, 35, 40, and 45 min [31, 33]. A non-irradiated sample was taken as a control. The treated seeds were then discharged in fixed quantities via a rotary valve, packed into polyethylene plastic bags, and stored at ambient air (29.9 °C and 64–66% RH) for 30 days before assessments.

Fungal and microbial count

Mold prevalence (cfu/g of grains) was determined according to the method outlined in [34] and [35]. Before being digested for 2 min, 25 g of representative samples were soaked for 30 min in 250 ml of sterile peptone (0.1% in water). 1 mL of the sample was serially diluted in 9 mL of peptone water, and 100 μL of the serial dilution was drop-plated on dichloron glycerol-18 (DG-18) agar medium (Oxoid chemicals, Hampshire, UK) for 4–5 days at 35 °C. The number of colonies (cfu/g) was recorded after incubation.

Germination percentage

100 g of seeds were germinated in Petri dishes (12 cm diameter) containing filter paper with 15 mL of distilled water. The seeds were considered to have germinated after radicle emergence. The germination period was 10–15 days, and the germination test ended when no seeds had germinated for 3 days. Germination percentage was calculated according to Sadeghianfar et al. [36] using the following formula: percentage of germination = (Total germinated seeds)/(Total number of seeds) × 100.

Respiration rate

The respiration rate was determined using the titration method described by Haney et al. [37]. 100 g of germinated seeds were placed in a desiccator with air free from CO_2 and a tube of 25 mL of 1 M KOH. After 1 h, KOH was titrated with 1 N HCl using a thymol blue indicator, and CO_2 production was calculated as mL CO_2 kg^{-1} seed h^{-1}.

Total phenolic content

Total phenolic content was determined according to Mohdaly et al. [38] as follows: the prepared ground sample (5 g) was macerated in 50 mL of each solvent (methanol 80%, ethanol 80%, and water) for 24 h at room temperature. Then, the crude solvent extracts were filtered through (Whatman No. 1) filter paper. Finally, filtrates were evaporated under a vacuum in a rotary evaporator at 45 °C and weighed to determine the extracted yield of each sample.

Total phenolic content was determined in each extract using Folin–Ciocalteu's method. An aliquot of the extract (0.1 mL) was mixed with 5 mL of distilled water and 0.5 mL of Folin–Ciocalteu's reagent, followed by one mL of Na_2CO_3 solution. The absorbance was measured after 2 h at 760 nm against a reagent blank. Chlorogenic acid was served as a standard compound and used to prepare the calibration curve in the range of 0–10 ppm. Phenolic content was expressed by mg Chlorogenic acid/100 g dry weight.

Essential oil

The oil was extracted by a hydro-distillation method in a Clevenger apparatus according to the Egyptian Pharmacopoeia [39] to determine the essential oil % (v/w). Seeds (100 g) were ground and immediately hydro-distilled for an average duration of 3 h.

Enzyme's activity

For the activities of peroxidase (POD), polyphenol oxidase (PPO), and catalase (CAT), enzyme extractions were conducted according to Yin et al. [40]. 5 g of powdered seeds were homogenized in 10 mL of 100 mmol K_2HPO_4 (pH 7.8) containing 1 mM EDTA and 1% (w/v) polyvinylpyrrolidone (PVP). The homogenate was centrifuged at 4 °C for 20 min at 12,000 rpm.

The enzyme activities were measured spectrophotometrically in the supernatant. For POD activity, 0.15 mL of enzyme extract was added to a reaction mixture containing 3 mL of 0.05 mol L^{-1} phosphate buffer (pH 7.8) and 0.04 mol L^{-1} guaiacol [41]. POD was measured as changes in the absorbance at 460 nm, and the results were expressed as U mg^{-1} protein. For PPO, the change in absorbance of a mixture containing 0.5 mL supernatant, 0.1 mol L^{-1} phosphate-buffered, and 0.5 mol L^{-1} catechol was obtained at 398 nm. PPO activity was expressed as U mg^{-1} [42]. CAT activity was determined according to Ravindran et al. [43]; an aliquot of 1 mL of the enzyme extract was added to the reaction mixture containing 3 ml of 0.1 M sodium phosphate buffer (pH 6.8) and 1 mL of 0.01 M H_2O_2. A decrease in H_2O_2 was monitored at 240 nm and considered enzyme activity. One unit of the enzyme was defined as μmol H_2O_2 decomposed per minute per mg protein (U mg^{-1}).

Statistical analysis

UV-C treatments were carried out in three replicates. For the statistical analysis, a one-way analysis of variance (ANOVA) was performed using SPSS (Version 26, IBM Corporation, USA), and the means of treatments were

Table 1 Germination percentage of anise, fennel, caraway, and cumin stored for 30 days after different UV-C exposure durations

Exposure time (min)	Anise	Fennel	Caraway	Cumin
0	48.30 ± 0.57 f	53.66 ± 1.52 e	52.33 ± 0.57 d	46.00 ± 2.00 f
5	50.00 ± 1.00 ef	56.66 ± 0.57 de	55.66 ± 0.57 cd	48.00 ± 1.10 ef
10	52.67 ± 0.57 de	57.00 ± 1.00 de	58.33 ± 0.57 c	48.66 ± 0.56 de
15	53.66 ± 1.52 cd	59.0 ± 1.10 cd	58.33 ± 1.52 c	50.66 ± 2.08 d
20	55.00 ± 2.00 cd	61.66 ± 1.52 bc	64.00 ± 1.20 b	51.00 ± 1.73 cd
25	67.66 ± 1.15 a	70.00 ± 1.3.00 a	69.33 ± 1.15 a	57.66 ± 2.08 a
30	65.33 ± 0.57 ab	64.66 ± 1.42 b	66.00 ± 1.00 ab	54.33 ± 0.67 b
35	63.66 ± 0.57 b	64.33 ± 0.47 b	65.66 ± 2.08 ab	53.00 ± 2.64 bc
40	62.33 ± 0.59 b	63.66 ± 1.15 b	65.00 ± 2.64 ab	53.33 ± 1.52 b
45	56.66 ± 1.52 c	62.66 ± 0.80 b	64.33 ± 1.54 b	52.66 ± 1.42 bcd

Data were expressed by mean ± standard deviation (SD); means within columns followed by the different lowercase letters indicate significant differences ($P \leq 0.05$). HSD at a 5% significance level was 3.25, 3.46, 4.42, and 2.1 for anise, fennel, caraway, and cumin, respectively

separated using Tukey's honest significance test (HSD) at a 5% significance level. Hierarchical cluster analysis (HCA) was performed to classify the effect of the different ultraviolet dosages based on the similarities among the measured variables by applying the squared Euclidean distance and Ward's linkage methods.

Results and discussion

Fungal and microbial count

The findings show that for all treated seeds, the total fungal and bacterial counts were around 4.1 and 1.67 × 10^2 cfu/g, respectively. Meanwhile, the control's total fungal and bacterial counts were 3.3 and 2.4 × 10^2 cfu/g, respectively, without significant differences between treated and untreated seeds. According to the data, all tested seeds had a lower microbiological load than those indicated by the Egyptian Specification Standards (ES: 1930/2008 and ES: 2095/2005), and the International Standards Organization (ISO: 9301/2003 and ISO: 2255/1996). This is because the spice legislation's microbial specifications set maximum limits of 10^4 and 10^2 CFU of total microbial and mold and yeast counts per gram, respectively [34, 35].

It's possible that the lower load in treated and untreated seeds is due to contamination in the field rather than storage, as 30 days of storage may not be long enough for an infection to become apparent, particularly following UV-C sterilization.

Germination percentage

The effect of different UV-C dosages on seed germination following a dormancy period of 30 days was examined. For all seeds, control samples had the lowest significant germination percentage of 48, 53, 52, and 46% for anise, fennel, caraway, and cumin, respectively. While the 25 min irradiated sample had the highest significant percentages of 67%, 70%, 69%, and 57%, respectively, as shown in Table 1. The germination percentage gradually decreased in the samples subjected to higher exposure periods. Similar results were found in Bengal gram (*Cicer arietinum* L.) and horse gram (*Macrotyloma uniflorum* L.) by Gandhi [44], who observed that germination percentage decreased gradually with a progressive increase in exposure duration of UV-C light. According to de Araujo et al. [45], UV-C up to a dose of 20.7 kJ m^{-2} resulted in a 50–66% increase in germination in castor oil plant seeds. In comparison, higher doses of 41.4 kJ m^{-2} reduced seed energy and decreased seed germination. According to Liu et al. [46], this reduction in the germination percentage suggested that energy was shifted to more functional metabolites to adapt in response to UV radiation.

Many investigations reported different impacts of UV on germination. For example, long-term UV-B radiation severely hampered the growth of *Astragalus mongholicus* seedlings [46] and inhibited barley seedlings' growth [28]. Whereas UV-C stimulated wheat germination, and increasing the exposure duration enhanced the germination percentage compared to the control [47]. Another study reported that UV-C had no significant effect on the germination percentage of maize and sugar beet seeds compared to the control [36]. HIDAKA and KUBOTA [48] reported that UV irradiation did not reduce germination compared to control.

According to Kovács and Keresztes [49], the biological effect of UV-C light (100–280 nm) as non-ionizing radiation is owing to photons with enough energy to break chemical bonds, creating a photochemical reaction as compared to visible light photons (> 400 nm). Furthermore, three primary mechanisms may have contributed

Table 2 Respiration rate (ml CO_2 Kg^{-1} seed h^{-1}) of anise, fennel, caraway, and cumin stored for 30 days after different UV-C exposure durations

Exposure time (min)	Anise	Fennel	Caraway	Cumin
0	24.46 ± 1.63 d	20.92 ± 1.76 f	20.33 ± 3.51 e	21.00 ± 1.73 h
5	26.92 ± 1.63 cd	21.22 ± 1.00 f	25.66 ± 2.08 de	27.33 ± 1.52 g
10	28.98 ± 2.22 bcd	22.73 ± 2.80 ef	25.66 ± 2.08 de	29.66 ± 1.52 fg
15	29.5 ± 2.40 bcd	24.13 ± 0.91 ef	27.00 ± 1.00 cd	33.00 ± 2.64 ef
20	30.33 ± 3.70 bc	25.62 ± 2.24 de	29.66 ± 2.08 cd	35.60 ± 1.15 de
25	42.15 ± 1.97 a	41.99 ± 3.30 a	46.33 ± 2.51 a	45.33 ± 0.57 a
30	38.80 ± 1.90 a	39.77 ± 1.79 a	40.33 ± 1.52 b	42.00 ± 2.00 ab
35	33.20 ± 3.20 b	33.90 ± 1.58 b	29.33 ± 4.16 cd	40.33 ± 1.52 bc
40	33.23 ± 3.02 b	30.62 ± 1.69 bc	31.66 ± 2.88 c	37.66 ± 1.15 cd
45	33.27 ± 3.60 b	29.36 ± 0.93 cd	28.33 ± 1.52 cd	36.33 ± 2.51 cde

Data were expressed by mean ± standard deviation (SD); means within columns followed by the different lowercase letters indicate significant differences ($P \leq 0.05$). HSD at a 5% significance level was 5.1, 4.3, 5.8, and 4.1 for anise, fennel, caraway, and cumin, respectively

Table 3 Total phenolic content (mg chlorogenic acid/100 g) of anise, fennel, caraway, and cumin stored for 30 days after different UV-C exposure durations

Exposure time (min)	Anise	Fennel	Caraway	Cumin
0	216.0 ± 2.1 ef	114.5 ± 0.9 c	26.0 ± 0.6 d	5.75 ± 0.35 h
5	213.5 ± 1.4 f	114.5 ± 0.7 c	26.0 ± 1.1 d	4.70 ± 0.28 h
10	221.5 ± 2.3 def	121.5 ± 1.1 c	31.5 ± 1.3 d	9.75 ± 0.35 ef
15	227.0 ± 2.8 de	119.5 ± 0.5 c	42.5 ± 0.5 c	11.25 ± 1.06 bc
20	245.0 ± 1.4 b	134.0 ± 1.7 b	54.0 ± 0.4 b	10.35 ± 0.91 ce
25	257.0 ± 0.7 a	145.0 ± 0.95 a	60.0 ± 0.7 ab	12.75 ± 0.35 a
30	240.0 ± 2.8 bc	143.0 ± 1.3 a	63.5 ± 0.8 a	10.0 ± 0.0 ce
35	244.0 ± 2.8 b	132.5 ± 0.4 b	62.5 ± 0.9 a	12.0 ± 2.82 ab
40	232.0 ± 2.8 cd	135.0 ± 1.6 b	54.0 ± 1.1 b	8.50 ± 0.70 fg
45	205.8 ± 1.4 g	120.0 ± 0.2 c	41.5 ± 1.3 c	7.50 ± 0.70 g

Data were expressed by mean ± standard deviation (SD); means within columns followed by the different lowercase letters indicate significant differences ($P \leq 0.05$). HSD at a 5% significance level was 11.17, 7.65, 8.31, and 1.4 for anise, fennel, caraway, and cumin, respectively

Table 4 Essential oil content % (v/w) of anise, fennel, caraway, and cumin stored for 30 days after different UV-C exposure durations

Exposure time (min)	Anise	Fennel	Caraway	Cumin
0	1.81 ± 0.05 d	1.28 ± 0.08 e	1.54 ± 0.05 b	5.78 ± 0.04 e
5	2.54 ± 0.12 b	1.31 ± 0.09 e	1.56 ± 0.03 b	5.86 ± 0.012 e
10	2.60 ± 0.04 b	1.46 ± 0.02 d	1.56 ± 0.04 b	6.21 ± 0.18 d
15	2.58 ± 0.04 b	1.54 ± 0.02 cd	1.65 ± 0.09 b	6.48 ± 0.13 c
20	2.90 ± 0.04 a	1.62 ± 0.03 abc	1.88 ± 0.04 a	6.88 ± 0.08 a
25	2.98 ± 0.05 a	1.71 ± 0.04 a	1.82 ± 0.01 a	6.82 ± 0.12 a
30	2.28 ± 0.11 c	1.68 ± 0.05 ab	1.78 ± 0.04 a	6.74 ± 0.12 ab
35	2.37 ± 0.05 c	1.63 ± 0.09 abc	1.78 ± 0.04 a	6.74 ± 0.04 ab
40	2.26 ± 0.09 c	1.59 ± 0.02 abc	1.6 ± 0.02 b	6.58 ± 0.09 bc
45	2.16 ± 0.08 c	1.58 ± 0.01 bc	1.57 ± 0.04 b	6.45 ± 0.04 c

Data were expressed by mean ± standard deviation (SD); means within columns followed by the different lowercase letters indicate significant differences ($P \leq 0.05$). HSD at a 5% significance level was 0.16, 0.12, 0.12, and 0.22 for anise, fennel, caraway, and cumin, respectively

to UV-C radiation's beneficial effect on seeds: (1) UV-C radiation caused a breakdown in the seed coat, allowing the seeds to absorb more oxygen and water and lessening dormancy; (2) UV-C radiation raised the temperature to be suitable for germination; and (3) UV-C radiation increased seed respiration and mitochondrial activities [36, 45]. According to Sarghein et al. [50], UV radiation influences growth via phytohormones through photo destruction or enzymatic reactions. Overall, UV light has different impacts on different species and cultivars of the same species.

It has been shown that there was no statistically significant difference in the percentage of germination for caraway seeds treated for 25–40 min ($P > 0.05$). However, this may depend on how caraway seeds respond to UV radiation during adaptation.

Respiration rate

The impact of ultraviolet radiation on *Apiaceae* spices' respiration rate is presented in Table 2. The findings showed that the respiration rate varied significantly depending on the exposure duration and that the respiration rate followed the same pattern as the germination percentage, which is consistent with the findings of Dahal et al. [51], who found that the respiration rate has a consistent relationship with germination. According to Horbach et al. [52], seeds with low respiration levels do not germinate rapidly or entirely. The seeds' dormancy is broken due to their absorption of radiation and energy conversion into heat, necessitating an increase in respiration to accommodate the cell-structure processes [36, 45]. Moreover, seed respiration and oxygen consumption have been used as good indexes of seed vigor and the activation of germinative metabolic processes [53].

The data revealed that the 25 min UV-treated samples had the highest significant respiration rate values for all types of seeds. However, the control and lower UV-C exposure duration-treated samples had the lowest significant values. In addition, Wang et al. [28] found that UV-B long-term radiation affected barley seedlings' physiological and metabolic functions, resulting in a significant reduction in respiration rate, which is consistent with our findings that higher UV-C exposure durations induced a significant decrease in respiration rate.

Phenolic content

Phenolic chemicals are the most prevalent secondary metabolites formed to protect plants in response to abiotic and biotic stresses, such as UV light and pathogens attack [54]. In addition, the high content of total phenolic and antioxidant activity gives an excellent rationale for using plant sources for medical purposes. According to Papoutsis et al. [55], ultraviolet treatment endorses the antioxidant capacity and bioactive compounds of horticultural products.

The data in Table 3 revealed a gradual increase in the total phenolic content by increasing the exposure time from 0 to 25 min, then a gradual decrease from 35 to 45 min without a significant difference with control, following Papoutsis et al. [55], who found that UV-C at low doses increased the phenolic content of lemon pomace powder, and a dosage higher than 19 kJ m^{-2} resulted in a reduction of total phenols. Even though all spices belong

to the same family, there is a variation in their overall phenol concentration. The results showed that the highest total phenolic content was obtained in anis and fennel, while the lowest values were obtained in caraway and cumin, comparable to [56–58]. Even though all spices belong to the same family, there is a variation in their overall phenol concentration. This may be due to the presence of different amounts of sugars, carotenoids, or ascorbic acid, the duration of cultivation, geographical differences, or extraction methods, which may alter the content of phenols [59, 60].

At 25 min exposure time, anise, fennel, and cumin had the highest significant values of total phenols ($P < 0.05$) of 257, 145, and 12.75 mg/100 g, respectively, while for caraway, the highest significant value of 63.5 mg/100 g was observed after 30 min of exposure.

According to Alothman et al. [61], phenylalanine ammonia-lyase, the key enzyme responsible for synthesizing and accumulating phenolic compounds, is promoted after UV-C treatment as a defense mechanism against irradiation. Brown et al. [62] reported that the low dose of UV-C possibly stimulated the production of phytoalexins physiologically in cabbage seeds, thus inducing black rot resistance in plants. Conversely, exposure to higher doses of UV-C suppressed phenylalanine ammonia-lyase activity and increased storage rots. The reduction of phenolic content at higher exposure time may be attributed to decreasing some phenolic compounds, such as phenolic acids or flavonoids [55]. In addition, the structure of seed tissues was affected by UV treatment, facilitating the release of polyphenols from cell wall polysaccharides, and increasing the value of total phenolic content. With increasing exposure time, the action of polyphenol oxidase degrades some of the free polyphenols by oxidation, causing a decrease in the phenolic content value [63]. The results of the current study confirm this.

Essential oil

In Table 4, data revealed that the essential oil content in anise seeds ranged from 1.81% to 2.98%. According to Ullah et al. [64], anise essential oil content ranges from 2.7% to 3%, whereas according to Shojaii and Abdollahi Fard [65], aniseeds contain 1.5–5.0% essential oil. From the obtained data, all UV-C treatments induced a significant increase in anise oil quantity compared to the control. In addition, UV-C-treated samples for 20, and 25 min exhibited the highest values of oil content of 2.90% and 2.98%, respectively, without a significant difference.

Özel et al. [66] reported that fennel essential oil content varied between 3.5% and 6%; however, in our investigation, the control sample yielded the lowest significant value of 1.28%, while the 25 min treated sample yielded the highest significant value of 1.71%. This variation between results might be due to the cultivar and the environmental conditions (soil moisture, temperature, soil nutrients, etc.) influencing seed quality [67]. In addition, it was observed that exposure durations of 0 and 5 min exhibited a significant decrease in oil content compared to higher durations ($P < 0.05$).

For caraway seeds, the oil content ranged from 1.54% to 1.88%, which is comparable to the findings of Raal et al. [68], who studied the content of essential oil of 20 commercial caraway seeds obtained from different countries and found that the investigated samples yielded an average oil content of 0.6–5.4%. Statistical analysis revealed that, despite the minor differences between treatments, there is a significant difference between them. However, there were no differences in oil contents between the samples exposed to UV-C for 20, 25, 30, and 35 min, and this range of exposure duration had a beneficial influence on the oil amount when compared to the control and other durations.

Cumin seeds yielded essential oil in the range of 5.78–6.88%. According to Moraghebi [69], the essential oil varied between 1.84 up to 5.06%, whereas other investigations reported a range of 2.0–4.0% [70, 71]. The exposure durations of 0 and 5 min resulted in a significant decrease in oil content compared to the longer durations. Furthermore, the difference in oil content for treated samples between 20 and 35 min was negligible.

To our knowledge, not much research has been conducted on the influence of UV-C on the essential oil of medicinal seeds. However, according to Erdoğdu and Ekiz [72] and [73], UV-C treatment is classified as a non-thermal process with no significant effect on weight loss or color changes, and volatile oil in cumin and black pepper seeds, which is comparable to our findings.

Antioxidant enzymes

As a mechanism of seed decay resistance, seeds have enzyme-based biochemical defenses positioned on or near the seed's surface [74]. Peroxidase (POD) is a key enzyme in plant antioxidant defense as it engages in metabolic activities related to respiration and promotes the utilization of phenolic compounds as co-substrates [75]. Catalase (CAT) is also essential to preserve the redox equilibrium during oxidative stress, because it scavenges H_2O_2 produced in the peroxisome organelle by catalyzing the dismutation of two molecules of H_2O_2 into water and oxygen [76]. Whereas the enzyme polyphenol oxidase (PPO) oxidizes *o*-diphenolic compounds, enhancing the defense efficacy of phenolics by creating highly reactive *o*-quinones [77].

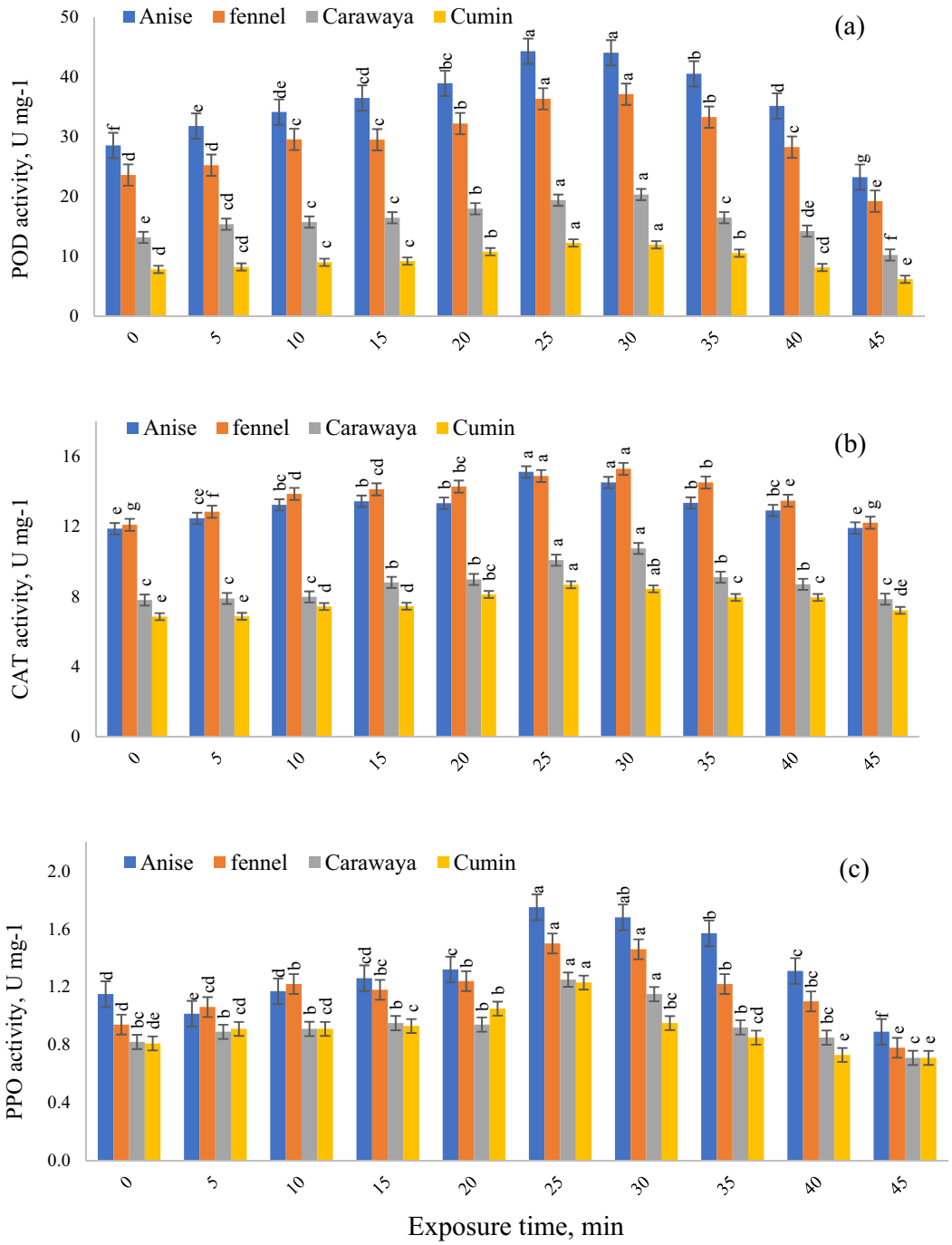

Fig. 1 POD (**a**), CAT (**b**), and PPO (**c**) activities in anise, fennel, caraway, and cumin stored for 30 days after different UV-C exposure durations. Different lowercase letters on top of the bars indicate significant differences ($P \leq 0.05$)

When compared to the untreated seeds, UV-C-treated seeds had a significantly higher POD, CAT, and PPO enzyme activity as the exposure time increased, as shown in Fig. 1. This is due to the development of antioxidant defenses against oxidative stress, as increased antioxidant enzyme activity scavenges excess reactive oxygen species. UV exposure has been known to stimulate the generation of reactive oxygen species (ROS), known as oxidative stress. Furthermore, at low levels of exposure, UV abiotic stressors promote the synthesis of secondary metabolites, such as carotenoids, phenolics, and flavonoids, as well as the development of antioxidant enzymes that

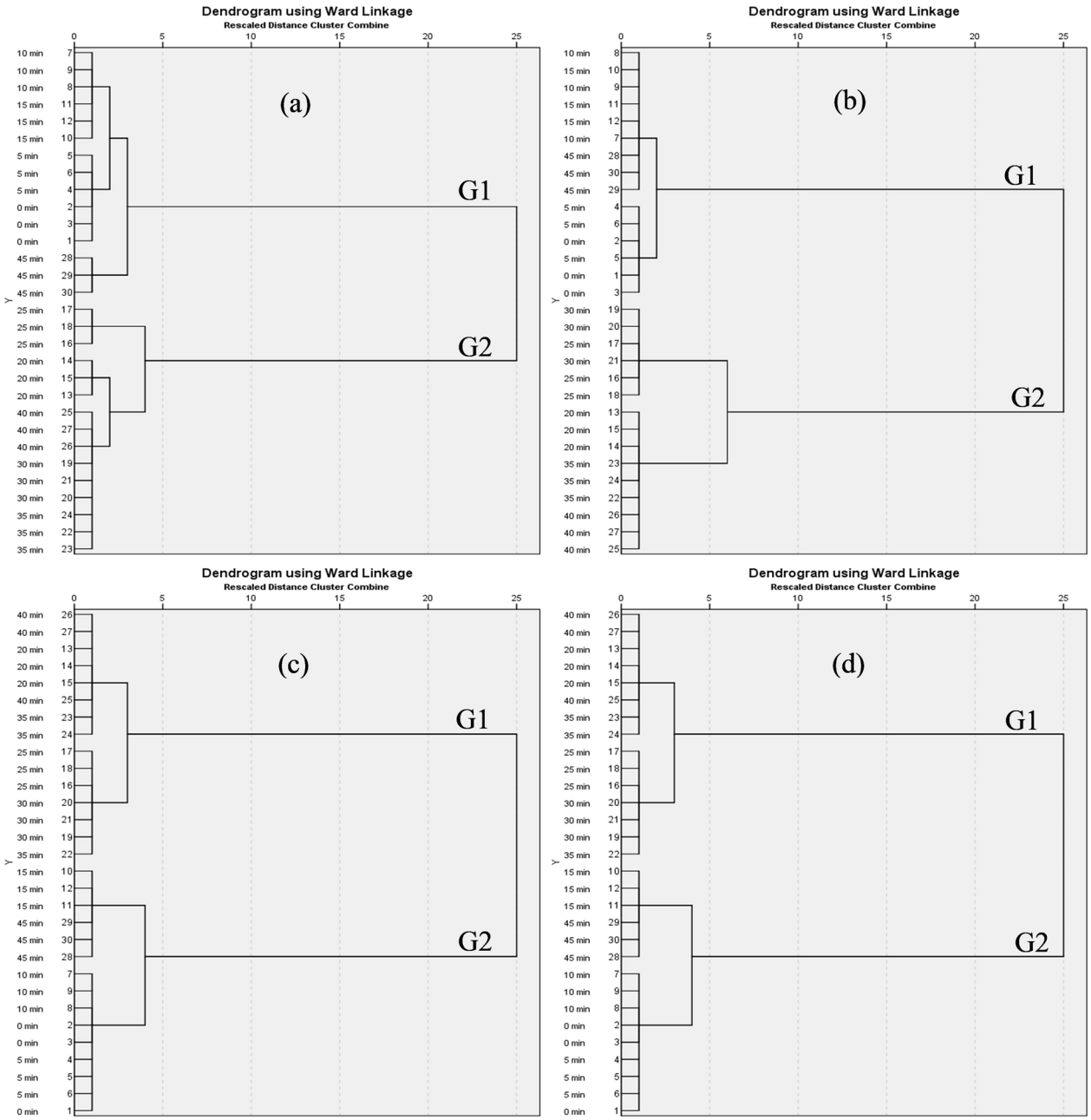

Fig. 2 Dendrogram exhibits the clustering according to similarities between the observations of all measured variables for **a** anise, fennel (**b**), caraway (**c**), and cumin (**d**) after the different durations of ultraviolet exposure. The measured variables were germination percentage, respiration rate, phenolic content, essential oil, and antioxidant enzymes

protect plants from oxidative damage, such as membrane lipid peroxidation [27, 30].

These results are comparable to those of Nasibi and M-Kalantari [27], they found that Brassica napus seedlings adapted to UV-B and UV-C as abiotic stressors by producing significantly more flavonoids and anthocyanins than the control. Furthermore, in response to UV-B and UV-C radiation, the activity of antioxidant enzymes increased in the leaves and roots of pepper plants in comparison with the control [75].

The results showed that POD activity progressively increased with the exposure durations, peaked at 25–30 min, and subsequently declined as the exposure time was extended. For example, after 25 min

of exposure, anise and cumin exhibited the highest increase in POD activity, approaching 155.2% and 156.6%, respectively. At the same time, fennel and caraway had the highest value of 157.2% and 154.5%, respectively, after 30 min of exposure, Fig. 1a. Meanwhile, 45 min of exposure caused a significant decrease in POD compared to the control. These results are consistent with the findings of Sharlaeva and Chirkova [30], who observed that POD activity increased in spring wheat seeds with increasing exposure durations, peaked at 30 min by 25.9%, and that a further increase in the time after 40 min resulted in a significant decline in POD compared to control.

The antioxidant mechanism is engaged in response to the UV-C stressor. With increasing irradiation time, POD activity increases, which could be attributed to the rise in reactive oxygen species synthesis, including hydrogen peroxide (H_2O_2) and lipid peroxidation. However, a further increase in UV exposure time reduces peroxidase activity significantly. According to Sharlaeva and Chirkova [30], this can be caused by a reduction in the intensity of hydrogen peroxide formation reactions and an increase in the metabolic processes that use H_2O_2, as well as the activation of the compensatory mechanisms that prevent the synthesis of free radicals, resulting in reduced enzyme synthesis.

According to the data in Fig. 1b, anise and cumin seeds treated for 25 min had the highest significant increase in CAT activity, reaching 127.3% and 126.6%, respectively. On the other hand, fennel and caraway seeds exposed for 30 min exhibited the highest significant increase of 126.4% and 137.82%, respectively. However, CAT activity was not significantly different at exposure time between 25 and 30 min. Furthermore, with increasing the exposure time, the activity of CAT gradually decreased to a level that was not significantly different from the control at 45 min of exposure time.

According to Costa et al. [29], UV-B radiation significantly boosted CAT activity in sunflower cotyledons seeds by 20% compared to the control. Moreover, UV-B radiation resulted in a CAT increase in rice seeds by 33–45% [78]. These findings suggested that CAT activity is crucial for controlling the amount of endogenous H_2O_2 produced in response to abiotic stressors, such as UV-C.

According to Flurkey and Inlow [79], Increased PPO in plant tissues in response to wounding suggests that PPO is produced as part of the defensive mechanism in plants. The data presented in Fig. 1c show that PPO activity in anise seeds increased significantly after 25 min of UV-C treatment, reaching 1.75 U mg^{-1} by 52.2%, and then reduced significantly with longer exposure times, down to 0.89 U mg^{-1} with 22.6% decrease after 45 min of exposure compared to control. The findings demonstrated that PPO activity in the other seeds followed the same pattern as in anise seeds. The UV-C radiation resulted in a maximum increase in PPO activity in fennel, caraway, and cumin by 59.6%, 52.4%, and 51.8%, respectively, at 25 min of exposure. Moreover, the data showed no discernible change in PPO among most other UV-C treatments, and there was no statistically significant difference between them.

Hierarchical cluster analysis (HCA)

The HCA was performed to differentiate between the various durations of ultraviolet exposure to identify the optimal dose that maintained the quality of seeds after 30 days of storage [80].

The data showed that the Dendrogram of anise seeds formed a small group of 25 min that represented the better quality among the six formed groups, Fig. 2a. Whereas for the higher quality in fennel, caraway, and cumin seeds, a group of 25–30 min duration was created and distinguished from the other durations groups, as shown in Fig. 2b–d. Furthermore, for fewer clusters in all seed types, two large groups were formed, one (G1) with intermediate durations of 20–40 min, and the other (G2) made up of shorter durations of 0–15 min and longer duration of 45 min, which is responsible for the reduced quality effect.

Conclusion

According to the results of the current study, low levels of UV-C light stimulate germination and respiration rate of *Apiaceae* spices as reliable indicators of a seed's vitality and the initiation of metabolic activities. The 25 min exposure had the highest significant values compared to the control. Moreover, as a defensive strategy against radiation, UV-C exposure encouraged an increase in phenolic content and essential oil in the examined seeds in the range of 20–35 min. Conversely, higher UV-C exposure caused a significant reduction in phenolic and oil contents. Antioxidant pathways play a crucial role in stress tolerance development and the increase in antioxidant enzymes in terms of POD, CAT, and PPO, owing to the development of protective mechanisms against oxidative stress. However, the decline in enzyme activity at higher UV doses of more than 25–30 min might be due to the stress caused by long-term UV exposure, which can have a devastating impact on cellular structures.

Storage of *Apiaceae* spices for extended periods in various packages under varied storage conditions is required to validate the influence of the acquired optimal dose of UV-C on their quality for consumption. A microbiological assay and additional biochemical parameter measures are needed in this situation.

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Author contributions
ME and RK: conceptualization, procedure, investigation, project administration, review. RK, NT, AE, and AA: methodology. LA and RK: data curation and statistical analysis, writing—original draft preparation, review, and editing. All authors read and approved the final manuscript.

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Availability of data and materials
The data sets used and/or analyzed during the current study are available to readers as in the manuscript and from the corresponding author upon reasonable request.

Declarations

Ethics approval and consent to participate
Not applicable.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

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