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RESEARCH ARTICLE

Chemical composition and antioxidant properties of γ-irradiated Iranian *Zataria multiflora* extracts

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Abstract

Context: Irradiation is the process of exposing food such as herbal plant to ionizing radiation to destroy microorganisms. *Zataria multiflora* Boiss (Lamiaceae), known as Avishan-e-Shirazi in Persian, is a thyme-like plant that grows naturally in central and southern parts of Iran and is used in traditional folk medicine.

Objective: In this study, the effects of γ-radiation on chemical composition and antioxidant properties of *Z. multiflora* were investigated.

Materials and methods: The plants were first irradiated with Co^{60} source (0, 10, and 25 kGy) and then subjected to Clevenger extraction to obtain essential oils. The composition of the oil was analyzed by a gas chromatography and compared with samples pretreated under different conditions. In parallel, the hydroalcoholic extract was prepared and used for measuring flavonoid content. Thereafter, the free-radical scavenging and antioxidant properties of essential oils and hydroalcoholic extract were examined.

Results: Despite the minor change in the individual oil constituents, the total percentage of the main components remained unaffected before and after irradiation (~95%). In addition, the total flavonoid content of hydroalcoholic extract was also unchanged due to irradiation (~32 mg QE/g extract). The high radical scavenging activity of the oil (~67%) and hydroalcoholic extract (~71%), in addition, the antioxidant properties of the oil (~91%) and hydroalcoholic extract (~95%), were unaffected after irradiation.

Discussion and conclusions: These findings may suggest the sustainability of *Z. multiflora* extract properties pretreated with γ-radiation. With a view to its antioxidant applications, resistance of *Z. multiflora* and its properties against radiation effects are promising findings.

Keywords: Essential oils, flavonoid, free-radical scavenging

Introduction

Irradiation of dried foods, particularly herbs and spices, has a great application potential and has already been implemented in many countries (Luckman, 2002). Many reports indicated that herbal plant and spices are susceptible to insect and disease attacks (Abou-Arab & Donia, 2001; Katuin-Raem et al., 2001), contamination by pesticides, heavy metals, and aflatoxins through different ways such as environment in developing countries, pollution in irrigation water, atmosphere, soil as well as sterilization methods and storage conditions (Abou-Arab et al., 1999; Evenhuis et al., 1995).

Irradiation treatment of food is becoming an increasingly accepted processing option for countries in the Asia-Pacific region wishing to meet growing sanitary and phytosanitary requirements in international trade (Luckman, 2002). The amount of spices irradiated commercially has been estimated in 2002 to reach about 100,000 tons worldwide. The advantages of irradiation of different food and food products are well established. The first application of irradiation is to reduce the number of pathogenic and spoilage microorganism (Farkas, 2011). The antimicrobial effect of γ-irradiation has been established by routine examination of food products before

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and after treatments. γ-Irradiation, by reducing the load of microorganisms, can increase the shelf life of the products, which is important in food preservation technology. In Iran, various products including dried spices and herbs are routinely irradiated. The conventional dose of γ-irradiation applied is 10 kGy, but radiation doses up to 30 kGy have been authorized for decontamination of dried food and spices (Mahindru, 2005; Farkas, 1977). Our routine analysis indicated high microbial load in Iranian *Zataria multiflora* Boiss (Lamiaceae) plant (data not shown). Therefore, as an herbal plant, its decontamination by a safe method can be considered as an important goal in herbal drug processing.

Z. multiflora, known as Avishan-e-Shirazi in Persian, is a thyme-like plant that grows naturally in central and southern parts of Iran. In Iran, *Z. multiflora* is used in traditional folk remedies for its antiseptic, analgesic, and carminative (antiflatulence and intestine-soothing) properties. It is extensively used as flavor ingredients in a wide variety of food especially in yoghurt flavoring. The essential oil was found to contain thymol and carvacrol as the major components, which are antimicrobial and antifungal agents (Sharififar et al., 2007; Mahboubi & Bidgoli, 2010; Saei-Dehkordi et al., 2010; Gandomi et al., 2009). Aqueous and alcoholic extracts of *Z. multiflora* have been therapeutically used for relieving nociceptive pain (Hosseinzadeh et al., 2000; Ramezani et al., 2004) and recurrent aphthous stomatitis (Jafari et al., 2003), and for preventing growth of oral streptococci (Owlia et al., 2004), *Plasmodium falciparum* (Ziegler et al., 2004), and *Trichomonas vaginalis* (Abdollahy et al., 2004), as well as used as an insect repellent (Saleem et al., 2004).

The usefulness of γ-irradiation is well known, provided that the chemical composition and biochemical properties of food and food products are retained. Despite the routine use of γ-irradiation, our knowledge about the influence of radiation on the chemical composition and pharmacological properties of spices is not well documented. The aim of this study was to investigate the chemical composition of *Z. multiflora* extracts pretreated with γ-irradiation. Moreover, the effect of γ-irradiation on antioxidant properties of the oils and hydroalcoholic preparations has been investigated.

Materials and methods

Chemicals

Aluminum trichloride ($AlCl_3$), chloroform, ethanol, dimethylsulfoxide, ascorbic acid, and methanol were purchased from Merck, Frankfurt, Germany. Quercetin, 2,2-diphenylpicrylhydrazyl (DPPH), trolox, β-carotene, linoleic acid, Tween 40, and butylated hyroxytoluene (BHT) were obtained from Sigma -Aldrich, St. Louis, MO.

Plant materials and radiation treatments

Fresh *Z. multiflora* was collected in May 2010 from the Shiraz city, Iran. Dr. Younes Asri (Botanist) authenticated the plant materials from herbarium of Iranian botanical garden (TARI) (Voucher Number: 41754). The whole aerial parts of the plants were divided into three batches (50 g) and packed in heat-sealed polyethylene pouches, and they were then passed to a Co^{60} source for irradiation at two different doses (10 and 25 kGy) using a high-dose rate research irradiator (Co^{60} Gammacell 220; Canada) calibrated with Fricke standard dosimeter (Co60 Gammacell 220 [Atomic Energy of Canada Limited Radiochemical Company], Canada), which is installed in Radiation application research school of atomic energy organization of Iran. The dose was controlled by the exposure time of each container to the source. The temperature and dose rate for all the samples were 22–23°C and 0.37 Gy/sec, respectively. The dose range within the samples was ±20% of the actual dose. The control and irradiated samples were stored in plastic containers at room temperature (28–30°C) under a similar condition.

Oil extraction and analysis

Oil extraction from the aerial parts of nonirradiated and irradiated *Z. multiflora* was carried out using a Clevenger-type apparatus. The extraction was carried out for 2 h, and the oils were stored in dark glass bottles in a freezer until further use. The oil extraction yield from both the nonirradiated and irradiated *Z. multiflora* seeds was approximately 4% (w/w). Gas chromatography (GC) analysis was performed using a GC (9-A Shimadzu, Kyoto, Japan) equipped with a flame ionization detector. Quantitation was carried out on Euro Chrom 2000 software (KNAUER Company, Berlin, Germany) by the area normalization method. The analysis was carried out using a DB-5 fused-silica column (30 m × 0.25 mm, film thickness 0.25 μm) and a temperature program of 40–250°C at a rate of 4°C/min, injector temperature of 250°C, detector temperature of 265°C, and the carrier gas was helium (99.99%). The GC/MS unit consisted of a Varian-3400 GC coupled to a Saturn II ion trap detector. The column of GC/MS was the same as of the GC under the same conditions that the above analysis was carried out. The constituents were identified by comparing their mass spectra with those in the computer library and with the authentic standards.

Preparation of hydroalcoholic extract

A known amount (30 g) of the powder prepared from *Z. multiflora* before and after irradiation was mixed with 50-mL distilled water and 50-mL methanol at 70–80°C and maintained at 60°C for 24 h. The hydroalcoholic extract was filtered through a Whatman filter #4 (pore size, 20–25 μm). The filtrate was then freeze-dried for further use. The extraction yield for the hydroalcohol extracts derived from nonirradiated and irradiated *Z. multiflora* seeds was ~9% (w/w).

Estimation of total flavonoids

Total flavonoid content of the hydroalcoholic extract was determined using the Dowd method, which is adapted by Arvouet-Grand et al. (1994). Briefly, 5 mL of extract

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solution (1 mg/ mL) was mixed with an equal volume of 2% $AlCl_3$ prepared in methanol. The absorbance was recorded at 415 nm using Shimadzu UV-3100 spectrophotometer after 10 min against a blank sample. The blank sample was prepared by mixing 5 mL of the extract with 5 mL of methanol alone. Total flavonoids were determined using a standard curve prepared with quercetin (0–100 mg/L) as the standard. The mean of three readings was used to calculate the flavonoid content, which was finally expressed as mg quercetin equivalents (QE)/g of hydroalcoholic extract.

Radical-scavenging capacity (DPPH assay) of the oils

The hydrogen atom or electron donation abilities of the extracts and pure compounds were measured from the bleaching of the purple-colored methanol solution of 2,2-DPPH. This spectrophotometric assay uses the stable radical DPPH as reagent (Burits & Bucar, 2000; Cuendet et al., 1997). Among the different concentrations of essential oil preparations, 20% v/v (in methanol) was found suitable for DPPH test. Fifty microliters of the essential oils in methanol was added to 5 mL of DPPH solution (0.004% DPPH in methanol). Trolox (1 mM), a stable antioxidant, was used as a synthetic reference. After 30 min of incubation period at room temperature, the absorbance was read against the blank at 517 nm. The inhibitory effects of the extracts in percent (I%) was calculated by the following formula:

$$I\% = (A_{\text{blank}} - A_{\text{sample}} / A_{\text{blank}}) \times 100.$$

where, A_{blank} is the absorbance of the control reagent (containing all reagents except the test compound), and A_{sample} is the absorbance of the test compound. All the assays were carried out in triplicate.

Radical-scavenging capacity of hydroalcoholic extract

The radical scavenging activity of the *Z. multiflora* hydroalcoholic extract was measured spectrophotometrically using DPPH radical (Blois, 1958). In this assay, 2 mL of the hydroalcoholic extract (50 μg/mL) was added to equal volume of DPPH solution (125 μM in methanol). The solution was then mixed and incubated at 37°C in dark for 30 min. The decrease in absorbance of DPPH was recorded at 517 nm. A parallel experiment was performed in which *Z. multiflora* extract was replaced with vitamin C (5 μg/mL) and considered as a positive control. The inhibition percentage was calculated by comparing the absorbance of the blank and the samples as discussed.

β-Carotene-linoleic acid assay of the oils

The antioxidant activity of essential oils was determined using the β-carotene bleaching test (Taga et al., 1984). Approximately, 10 mg of β-carotene (type I synthetic) was dissolved in 10 mL of chloroform, and then, 0.2 mL of this solution was added to a boiling flask containing 20-mg linoleic acid and 200-mg Tween 40. Chloroform was removed using a rotary evaporator at 40°C for 10 min. Then, 50 mL of distilled water saturated with oxygen was added slowly with vigorous agitation to form an emulsion. The emulsion (5 mL) was added to a tube containing 0.2 mL of essential oil solution prepared according to Choi et al. (2000). The absorbance was immediately measured at 470 nm against a blank consisting of an emulsion without β-carotene. The tubes were placed in a water bath at 50°C and the oxidation of the emulsion was monitored spectrophotometrically by measuring absorbance at 470 nm over a 60-min period. Samples containing 0.2 mL of ethanol instead of essential oils were also monitored and used as control. Butylated hydroxytoluene (BHT; 1 mM in ethanol), a stable antioxidant, was used as reference. The antioxidant activity was expressed as inhibition percentage with reference to the control sample after 60 min of incubation, using the following equation: $AA = 100(DR_C - DR_S)/DR_C$, where

AA = antioxidant activity,
DR_C = degradation rate of control = [ln(a/b)/60],
DR_S = degradation rate in presence of sample = [ln(a/b)/60],
a = absorbance at time 0,
b = absorbance at 60 min.

β-Carotene-linoleic acid assay of hydroalcoholic extract

In this assay, antioxidant capacity was determined by measuring the inhibition of the volatile organic compounds and the conjugated diene hydroperoxides arising from linoleic acid oxidation (Dapkevicius et al., 1998). A stock solution of β-carotene–linoleic acid mixture was prepared as follows: 5-mg β-carotene was dissolved in 10 mL of chloroform (HPLC grade), and then, 25-μL linoleic acid together with 200-mg Tween 40 were added. Chloroform was completely evaporated using a vacuum evaporator before adding 100-mL distilled water saturated with oxygen. After dissolving the residue, an aliquot (5 mL) of this mixture was dispensed into a test tube. Then, 350-μL of the *Z. multiflora* extract, prepared at 2 g/L concentrations, was added, and the emulsion system was incubated for 2 h in a water bath at 50°C. Assay containing BHT (2 g/L) was also carried out and considered as positive control. After the incubation period, the absorbance was recorded at 470 nm. Antioxidative capacity of the hydroalcoholic extract was compared with those of BHT and the blank.

Statistical analysis

Data are presented as means ± standard error. The results were subjected to one-way analysis of variance followed by Tukey's Honestly Significant Differences using SPSS13.0 software. The significance was considered as $p < 0.05$.

Results and discussion

Effect of γ-irradiation on chemical composition and antioxidant properties of *Z. multiflora* essential oils

GC/GC-MS analysis of essential oil extracted from aerial parts of *Z. multiflora* cultivated in Shiraz city,

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Table 1. Chemical composition of essential oils from *Zataria multiflora* pretreated with γ-irradiation.

		Control		γ-Irradiated at 10 kGy		γ-Irradiated at 25 kGy	
No	Compound	RI	%	RI	%	RI	%
1	α-Thujene	931.8698	0.3937	931.8698	0.5731	931.8698	0.7309
2	α-Pinene	940.7259	2.3912	940.7259	3.5256	941.8157	4.7027
3	β-Pinene	976.7905	1.0692	976.7905	2.0814	976.7905	2.359
4	α- Terpinene	1031.1512	0.7535	1031.1512	1.554	1032.5203	1.7354
5	P-Cymene	1039.3074	7.5785	1040.6534	16.2099	1041.9956	17.5439
6	γ-Terpinene	1073.1315	4.4083	1074.386	8.6285	1074.386	9.3168
7	Thymol	1306.5475	61.8047	1309.15	49.3027	1307.8499	45.353
8	Carvacrol	1315.6156	10.5496	1316.9019	6.6025	1316.9019	6.3301
9	Thymyl acetate	1358.113	2.8072	1359.3259	3.033	1359.3259	3.1864
10	Geramyl acetate	1373.7243	0.4918	1373.7243	0.346	1373.7243	0.3911
11	E-Caryophyllene	1458.1412	2.581	1458.1412	2.7041	1458.1412	2.6727
12	Aromadendrene	1476.2953	0.9966	1476.2953	1.1283	1476.2953	1.1539

GC analysis was performed by a GC (9-A-Shimadzu) gas chromatograph equipped with a flame ionization detector. The constituents of the *Z. multiflora* oil extracts were identified by comparison of their mass spectra with those in the computer library and with authentic standards.
GC, gas chromatography; RI, retention index.

Iran, resulted in identification of 12 known compounds (Table 1). The major compounds were thymol (61.8%), carvacrol (10.5%), *p*-cymene (7.5%), and γ-terpinene (4.4%). Similarly, other studies also indicated thymol and carvacrol as the main constituents of this plant (Ali et al., 2000; Shaffiee & Javidnia, 1997; Basti et al., 2007; Sharififar et al., 2007; Mahboubi & Bidgoli, 2010). Characterization of the chemical composition of essential oils in *Z. multiflora* from different parts of Iran also indicated that thymol, a phenolic compound of oxygenated monoterpens, was the most abundant component in GC/MS ranging from 27.05% to 64.87% with the high antioxidant activities (Saei-Dehkordi et al., 2010).

Comparison of the essential oil fractions in the *Z. multiflora* before and after γ-irradiation (10 and 25 kGy) analyzed by GC/MS showed minor changes in the total percentage of oil chemical constituent (Table 1). These changes were mainly recorded in thymol (–16.5%), carvacrol (–4.2%), p-cymene (+10%), and γ-terpinene (+4.9%) irradiated at 25 kGy. Irradiation at 10 kGy caused small changes in the level of some essential oils, particularly thymol (–12.5%), carvacrol (–3.9%), *p*-cymene (+8.7%), and γ-terpinene (+4.2%). These data were further supported by showing equal oil yields of approximately 4% (w/w) in irradiated and control seeds.

The antioxidative properties of the *Z. multiflora* essential oils before and after γ-irradiation (10 and 25 kGy) analyzed by DPPH and β-carotene bleaching tests are presented in Table 2. When compared with a standard antioxidant agent, that is, Trolox (15%), it was found that essential oils extracted from *Z. multiflora* have strong radical scavenging activity (67%). Addition of the plant oils to the reaction mixture containing β-carotene and linoleic acid, brought about ~91% inhibition in formation of peroxidation products that was maintained even after irradiation (Table 2). The DPPH assay data revealed that γ-irradiation at lower dose (10 kGy), which is routinely used for food preservation, and also sterile dose (25 kGy) cannot influence the DPPH radical scavenging capacity of the oils ($p > 0.05$).

Table 2. Free radical scavenging and antioxidant activities (%) of *Zataria multiflora* essential oils.

Antioxidant	β-Carotene	DPPH
Nonirradiated essential oil	90.56 ± 0.25	67.05 ± 0.12
Irradiated essential oil (10 kGy)	92.16 ± 1.79	66.96 ± 0.04
Irradiated essential oil (25 kGy)	92.39 ± 0.76	66.81 ± 1.63
Trolox	—	15.6 ± 0
BHT	65.98 ± 1.21	—

The radical scavenging activity of the oil samples were measured spectrophotometrically using DPPH free radical. Trolox was used as positive control. The antioxidant activity of the *Z. multiflora* oil samples were measured spectrophotometrically using β-carotene bleaching test. Butylated hyroxytoluene was used as positive control. Results are mean ± SEM of three analyses carried out on essential oils derived from fresh and irradiated *Z. multiflora* Boiss.

In regard to these data, we can demonstrate that the unaffected radical scavenging and antioxidant properties of the oils can be attributed to the unchanged total oil components due to γ-irradiation obtained by GC/MS analysis. Despite changes in the level of some individual oil constituents due to irradiation, the total percentage of the main components, that is, thymol, carvacrol, *p*-cymene, and γ-terpinene remained unaffected before (95.82%) and after irradiation at 10 and 25 kGy (95.69% and 95.47%, respectively). Nevertheless, irradiation caused decrease in thymol and carvacrol levels and increase in γ-terpinene and *p*-cymene percentages. Studies indicated that all of these constituents possessed antioxidant activities, which compensate the individual oil decreasing (Mechergui et al., 2010; Celik et al., 2010). This was confirmed by experiments using γ-terpinene as an inhibitory factor in the peroxidation process of linoleic acid, which facilitated cross-reactions between HOO° and LOO° radicals leading to rapid chain

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termination of the peroxidation pathway (Foti & Ingold, 2003). One study showed that the phenolic -OH groups present in oil of thymol, which act as hydrogen donors to the peroxy radicals produced during the first step of lipid oxidation, is probably responsible for retardation of hydroxyl peroxide formation (Farag et al., 1989). Teissedre and Waterhouse (2000) also reported a good correlation ($r = 0.75$) between the total phenol content of essential oils containing thymol, carvacrol, and *p*-cymene and human low-density lipoprotein oxidation *in vitro* (Teissedre & Waterhouse, 2000). Our previous studies on caraway seeds also indicated that γ-irradiation could not influence the components and also antioxidant properties of the seeds extracts *in vivo* and *in vitro* systems (Fatemi et al., 2010a,b & 2011). Also, complete decontamination of peppermint after low-dose γ-irradiation (0.5, 1.0, and 2.66 kGy) did not lead to significant loss in essential oil components (Machhour et al., 2011).

Effect of γ-irradiation on total flavonoid content and antioxidant properties of the *Z. multiflora* hydroalcoholic extracts

The total flavonoid content of *Z. multiflora* was found to be ~32 mg of quercetin (QE)/g hydroalcoholic extract, which was significantly unaffected in samples preexposed to 10 and 25 kGy radiation ($p > 0.05$) (Figure 1). However, the extraction yields of hydroalcoholic extracts after γ-irradiation remained at the same level (~9% w/w) in nonirradiated sample. The antioxidant properties of hydroalcoholic extracts derived from *Z. multiflora* assessed by the DPPH and β-carotene bleaching test indicates strong antioxidant ability of the extract. In comparison to the standard antioxidant agent, that is, vitamin C (~48%), it was found that *Z. multiflora* hydroalcoholic extract possessed stronger radical scavenging activity (~71%) (Table 3). In β-carotene bleaching test, it was also found that *Z. multiflora* hydroalcoholic extract caused ~95% inhibition in formation of peroxidation products (Table 3). The antilipid peroxidation activities of hydroalcoholic extracts from other natural sources have also been reported (Tepe et al., 2005). One study also indicated that sub-fractions of the methanol extract of *Z. multiflora* were able to reduce the stable free radical 2,2-DPPH with the high inhibition values of linoleic oxidation (Sharififar et al., 2007). The results presented in Table 3 shows that γ-irradiation could not change the radical scavenging activity of the hydroalcoholic extracts ($p > 0.05$). As well, the extracts from γ-irradiated seeds failed to alter the lipid peroxidation reaction (β-carotene bleaching test) (Table 3). These results are in agreement with the report of Farag and el-Khawas (1998) showing the lack of influence of γ-irradiation on antioxidant property of caraway extracts. The antioxidant properties of sage, thyme, and oregano in chloroform and methanol extracts as well as in their mixture were also unaffected after irradiation at 10 kGy (Brandstetter et al., 2009). The result of one study indicated that γ-irradiation 0.5–10 kGy does not have adverse changes in biological activity of *Schizandra chinensis* Baillon (Schisandraceae) extract (Lee et al., 2011).

The lack of changing in antioxidant and DPPH scavenging activity due to irradiation were associated with the amount of unaffected flavonoid content in the irradiated *Z. multiflora* preparations (Figure 1). Our results together with reports from other laboratories suggest that the total flavonoid content of plant play a major role in antioxidant and radical scavenging activity of the hydroalcoholic extract (Jimoh et al., 2007; Jeong et al., 2007). It is likely that flavonoids with 3-OH group have a major contribution to the antioxidant activity of the natural products (Jeong et al., 2007). A correlation between the total flavonoid content and antioxidant activity was also reported in methanol extracts of *Paullinia pinnata* L. (Sapindaceae) leaves (Jimoh et al., 2007). One study also indicated that γ-radiation dose of up to 10 kGy was found to be sufficient for complete microbial decontamination without affecting the bioactive properties of herbal formulations, including antioxidant potential,

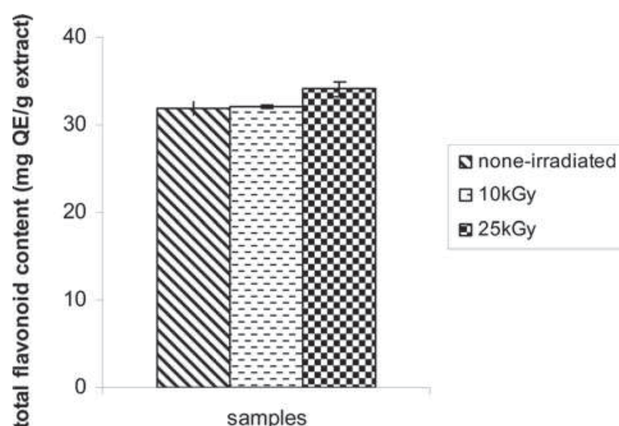

Figure 1. Effect of γ-irradiation on total flavonoid content of *Zataria multiflora*.

Table 3. Free radical scavenging and antioxidant activities (%) of *Zataria multiflora* hydroalcoholic extracts.

Antioxidant	β-Carotene	DPPH
Non-irradiated hydroalcoholic extract	95.54 ± 0.08	72.36 ± 7.69
Irradiated hydroalcoholic extract (10 kGy)	94.99 ± 1.39	64.75 ± 0.5
Irradiated hydroalcoholic extract (25 kGy)	96.98 ± 0.65	67.89 ± 0.64
Vitamin C	—	47.85 ± 0.28
BHT	97.4 ± 0.59	—

The radical scavenging activity of the hydroalcoholic extract prepared from *Z. multiflora* was measured spectrophotometrically using the DPPH free radical. Vitamin C was used as positive control. The antioxidant activity of the hydroalcoholic extracts prepared from *Z. multiflora* was measured spectrophotometrically using β-carotene bleaching test. BHT was used as positive control. Results are mean ± SEM of three analyses carried out on hydroalcoholic extracts derived from fresh and irradiated *Z. multiflora* Boiss.

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which was high in rasayan, shatpatryadi, scrub, rose, and guggul. The antioxidant property of these herbals could be attributed to components such as phenolics, flavonoids, and color pigments (Kumar et al., 2010). Another study also showed that γ-irradiation at 5 kGy could be a potential method to decontaminate the microbial load of *Polygoni multiflori* Radix without significant changes in its total phenols and antioxidant properties (Chiang et al., 2011). The 30 kGy dose applied to dry sage and oregano for sanitization did not significantly affect the capacity to inhibit the DPPH radical or the reducing power, nor did it affect the total phenolic content of the methanol and aqueous extract (Pérez et al., 2011).

Conclusions

These findings suggest sustainability of *Z. multiflora* extract properties on exposure to γ-radiation. With a view to its antioxidant applications, resistance of *Z. multiflora* and its properties against radiation effects are promising findings. The evidences presented in this article show that neither the chemical composition nor the *in vitro* antioxidant properties of the *Z. multiflora* extracts are affected by γ-irradiation. Nevertheless, it appears that there are positive effects on the *in vitro* antioxidant properties due to the conventional method of γ-irradiation used in food preservation.

Declaration of interest

The authors declared no conflict of interest.

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