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Post-harvest treatment of cherry tomatoes by gamma radiation: Microbial and physicochemical parameters evaluation



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ARTICLE INFO

Article history: Received 21 March 2016 Received in revised form 20 April 2016 Accepted 10 May 2016 Available online 20 May 2016

Keywords:
Cherry tomatoes
Gamma radiation
Microbial inactivation
Microbial food safety
Antioxidant activity
Physicochemical parameters

ABSTRACT

The aim of this study was to evaluate the effects of gamma radiation on cherry tomatoes, to assess the potential of irradiation post-harvest treatment for fruit shelf-life extension. Freshly packed cherry tomatoes (*Solanum lycopersicus* var. *cerasiforme*) were irradiated at several gamma radiation doses (0.8 kGy up to 5.7 kGy) in a ⁶⁰Co chamber. Microbiological parameters, antioxidant activity and quality properties such as texture, color, pH, total soluble solids content, titratable acidity, and sensory parameters, were assessed before and after irradiation and during storage time up to 14 days at 4 °C. Inactivation studies of natural cherry tomatoes microbiota and inoculated potential foodborne pathogens (*Salmonella enterica*; *Escherichia coli* and *Staphylococcus aureus*) were performed. A two log reduction on the microbial load of cherry tomatoes was verified after irradiation at 3.2 kGy, and 14 days of storage at 4 °C. Moreover, a maximum reduction of 11 log on the viability of potential foodborne bacteria was obtained after irradiation at 3.2 kGy on spiked fruits. Regarding fruits quality properties, irradiation caused a decrease in firmness compared with non-irradiated fruit, although it was verified a similar acceptability among fruits non-irradiated and irradiated at 3.2 kGy. Therefore, these results suggest that the irradiation treatment could be advantageous in improving microbial safety of cherry tomatoes and shelf-life extension without affecting significantly its quality attributes.

Industrial relevance: There is an ever-increasing global demand from consumers for high-quality foods with major emphasis placed on quality and safety attributes. One of the main demands that consumers display is for minimally processed, high-nutrition/low-energy natural foods with no or minimal chemical preservatives. Extending the shelf-life, while improving the food safety, will have a positive impact on both the industry and consumers (and potential target groups such as immunocompromised patients). The present study indicated that post-harvest gamma radiation treatment of cherry tomatoes can be used as an emergent, clean and environmental friendly process to extend the shelf-life of this fruit with safety and quality.

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1. Introduction

Tomato is an important agricultural commodity worldwide because of its contribution to human health and nutrition (Soto-Zamora, Yahia, Brecht & Gardea, 2005), and cherry tomato fruit is especially popular all over the world. The cherry type is a fresh tomato specialty having a small size, tomato-like flavor, and firm texture (Ergun, Sargent, & Huber, 2006). The shelf stability of this fruit ranges from five to seven days depending on the time of harvest, showing some limitations regarding fresh market utilization. Cherry tomato (*Solanum lycopersicus* var. *cerasiforme*) is a fruit present in diets worldwide, rich in antioxidants, like carotenoids, ascorbic acid and phenolic compounds (Raffo et al., 2002) that can promote beneficial effects when ingested.

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Tomatoes constitute the predominant source of lycopene in most diets, and this compound has been associated with a range of health benefits (Slimestad & Verheul, 2005). It is known that the consumption of cherry tomato may prevent several types of cancer in the digestive tract (La Vecchia, 1998).

During production, the fruit is exposed to conditions that are prone to contaminate the product. Such conditions include for instance irrigation waters, fields fertilized with manure, inappropriate seeding or sick workers (Heaton & Jones, 2008). Despite the risks, these products reach the consumers without an effective treatment process that may compromise their quality and shelf-life. Moreover, these fruits can host foodborne pathogens that may constitute a serious threat, causing gastrointestinal diseases when ingested. Also, food could serve as vectors for spread of such diseases when exported to another region (EFSA, 2012; Newell et al., 2010). In recent years, fresh tomatoes have attracted public attention due to the association with more than thousand

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reported outbreaks around the world (Canadian Food Inspection Agency, 2012; Center for Science in the Public Interest, 2010; Solano et al., 2013; Valadez, Schneider, & Danyluk, 2012). Consumers can wash the products to remove microorganisms, but even using disinfectants, the washing process has a limited success to remove deterioration microorganisms and pathogens (U.S. Food and Drug Administration, 2009).

To better control pathogenic contamination during the entire process, irradiation methods might represent the most effective method for decontamination with log reductions seen up to 7.0 for foodborne pathogens (Farkas & Mohácsi-Farkas, 2011; Goodburn & Wallace, 2013; Lynch, Tauxe, & Hedberg, 2009). The irradiation process also has the capacity to improve some nutritional properties, like an increase of the antioxidant activity (Cabo Verde et al., 2013). Radiation technologies have the ability to inactivate microorganisms without changing temperature. Therefore, it is possible to avoid deterioration of flavor, color and nutrient value of food as that induced by heat. Food irradiation can be performed after the final packaging stage, without any further intervention, reducing cross contamination, until it reaches consumers. Despite its development for the last 100 years (Molins, 2001), food irradiation technology is still having a slow implementation, mainly due to social and political factors (Farkas & Mohácsi-Farkas, 2011; Goodburn & Wallace, 2013; Jermann, Koutchma, Margas, Leadley, & Ros-Polski, 2015). Moreover, large scale adoption of this process for the decontamination of produce has not been taken up by the fresh produce industry. This could be due to the need for further research in food irradiation to evaluate the effects on fruits and vegetables of the radiation doses required for controlling several pathogenic organisms (Goodburn & Wallace, 2013).

Studies have been undertaken to reduce the microbial load on fresh fruits and vegetables, which include cherry tomatoes, using chemicals and other physical processes (Daş, Gürakan & Bayindirli, 2006; Song, Choi, & Song, 2011; Yun, Fan, & Li, 2013). Although, to the best of our knowledge, there is no study concerning the assessment of gamma radiation, as a clean and environmentally friendly technology, to reduce the load of natural microbiota and potential pathogenic bacteria on cherry tomatoes, considering its health-promoting and industrial significance.

The aim of this study was to evaluate the effects of gamma radiation on cherry tomatoes in order to access the potential use of gamma radiation as a post-harvest treatment process to further increase the safety, quality and economic value of this fruit.

2. Material and methods

2.1. Sampling and irradiation process

Cherry tomatoes (*Solanum lycopersicus* var. *cerasiforme*) with light red color from greenhouse production, were purchased (between April 2014 and February 2015) from a local market in Lisbon, Portugal and immediately kept at 4 ± 1 °C and transferred to the laboratory for experiment.

Polystyrene boxes containing approximately 125 g of fruits were irradiated at room temperature in a ^{60}Co experimental equipment (Precisa 22, Graviner, Lda, UK), with an activity of 165 TBq (4.45 kCi) and a dose rate of 1.8 kGy/h, located at the Campus Tecnológico e Nuclear, Bobadela, Portugal. The boxes containing the samples were irradiated at 1.3 kGy, 3.2 kGy and 5.7 kGy. The spiked cherry tomato samples with bacterial strains suspensions were irradiated at the doses ranging from 0.4 and 3.0 kGy for Salmonella Typhimurim and 0.77 to 1.22 kGy for Staphylococcus aureus and Escherichia coli. The dose rate was determined by Fricke dosimetry (America Society for Testing Materials, 1992). Absorbed doses were measured by routine dosimeters (Amber Perspex, Batch X, Harwell®, London, UK) with nominal uncertainty limits of about 2.5% (Whittaker & Watts, 2001). Three independent irradiation batches were performed. An average uniformity of dose (D_{max}/D_{min}) of 1.6 was obtained.

After irradiation the fruits in closed polystyrene boxes were kept under refrigerated conditions (in a freezer at 4 $^{\circ}$ C) until analysis. Microbiological and physicochemical parameters were evaluated after 0, 7 and 14 days of storage. Triplicate independent samples were used for each parameter, as well as non-irradiated samples (0 kGy) that followed all the procedures.

2.2. Microbial inactivation studies

2.2.1. Natural microbiota

Non-irradiated and irradiated cherry tomatoes (25 g) were placed in sterile stomacher bags containing 100 mL of 0.1% Tween 80 physiological solution. Samples (n = 9/dose) were homogenized using a stomacher (Stomacher 3500; Seaward, UK) for 15 min. Serial decimal dilutions were prepared for inoculation on Tryptic Soy Agar plates (TSA) (Oxoid LTD, Basingstoke, England) for bacterial counts and Malt Extract Agar plates (MEA) (Merck KGaA, Darmstadt, Germany) for filamentous fungi counts. Samples were incubated at 30 °C for TSA plates and 28 °C for MEA plates and colony numbers were counted for 7 days. The results were expressed as log colony-forming units (CFU) per gram of fresh fruit.

To evaluate the microbial stratification of cherry tomatoes, the skin was separated from the pulp of intact fruit samples using a sterile spoon, and both fruit parts were analyzed for bacterial and fungal counts as described previously.

All isolated colonies, from irradiated and non-irradiated samples, were characterized macroscopically (e.g., shape, pigmentation, texture), microscopically (e.g., cell shape on bacteria, morphology and soma in fungi), biochemically (gram staining, catalase activity, cytochrome oxidase) and organized in typing groups according Bergey's Manual of Determinative Bacteriology (Holt, Krieg, Sneath, Staley, & Williams, 1994). Afterwards, the frequency of each morphological group was calculated based on the number of each isolated type.

2.2.2. Artificial inoculation — challenging tests

Artificial contamination assays were carried out using three different bacterial strains in separated sets. Strains of Staphylococcus aureus (ATCC 6538), Salmonella enterica serotype Typhimurium (ATCC 14028) and Escherichia coli (ATCC 8739) were used in this study. Each of the strains was maintained at -80 °C in Tryptic Soy Broth (TSB; Merck KGaA, Darmstadt, Germany), with a 50% glycerol solution. Prior to use the bacterial strains were streaked on TSA and then incubated at 37 °C for 24 h. Suspensions of each microorganism were prepared in physiological solution. The concentration of the inoculums was approximately 8 log CFU/mL, as determined by serially diluting the inoculums and plating on TSA. These inoculums were used in subsequent experiments. Aliquots of the prepared bacterial suspensions were spot-inoculated onto the surface of 25 g of cherry tomatoes to achieve a concentration of 6 log CFU/g. The inoculums were let to dry (30 min) in a laminar-flow cabinet and the samples were placed in sterile stomacher bags for irradiation. The spiked non-irradiated and irradiated samples (n = 9 samples/dose and per bacterial strain) were analyzed for bacterial counts as described in the previous section using the selective media of Violet Red Bile Agar (Merck KGaA, Darmstadt, Germany) for E. coli; Xylose Lysine Deoxychloate Agar (Merck KGaA, Darmstadt, Germany) for S. Typhimurium and Baird Parker Agar (Merck KGaA, Darmstadt, Germany) for S. aureus. Plates were incubated at 37 °C for 7 days, and colonies were subsequently enumerated. The detection limit of the method was 1 CFU/g. The microbial counts were recorded and expressed as the log CFU/g.

2.3. Total phenolic content and antioxidant activity

The extraction method used was adapted from the described by Larrauri, Rupérez, and Saura-Calixto (1997). Samples of cherry tomato were previously irradiated as described in 2.1 and freeze-dried (Heto

PowerDry DW8 Freeze Dryer, Waltham, US) for 140 h. An amount of 0.5 g of the lyophilized samples were mixed with 20 mL of methanol, sonicated during 10 min and centrifuged for 5000 rpm, at 20 °C for 15 min (Beckman J2-21M Induction Drive Centrifuge, Ramsey, US). The supernatant was used in the following assays. The samples were maintained at 4 °C between assays.

Total Phenolic Content (TP) was determined based on Folin-Ciocalteau method (Singleton, Orthofer, & Lamuela-Raventós, 1998) using Gallic acid (Sigma, St. Louis, US) as standard for the calibration curve. The sample absorbance was read at 765 nm using a spectrophotometer (Shimadzu UV 1800, Kyoto, Japan). The results were expressed as mg of gallic acid equivalent (GAE) per 100 g cherry tomatoes fresh weight (fw). The assays were carried out in triplicate.

The antioxidant activity was determined by two procedures: DPPH assay, described by Brand-Williams, Cuvelier, and Berset (1995) and Ferric Reduction/Antioxidant Power (FRAP assay), described by Benzie and Strain (1996).

For DPPH method, samples aliquots of $100~\mu L$ at several concentrations were added to 3.9~mL of DPPH $60~\mu M$ and the decrease in absorbance was determined at 515~nm (Shimadzu UV 1800, Kyoto, Japan) at 0~min and every 30~min until the reaction reached a plateau. The results were expressed as EC_{50} which represents the concentration of extract necessary to decrease the initial DPPH concentration by 50%.

For FRAP assay, the reagent was freshly prepared by mixing 300 mM of acetate buffer (pH 3.6), 10 mM of 2,4,6-Tris(2-pyridyl)-s-triazine (TPTZ; Fluka, Buchs, Switzerland) and 20 mM FeCl₃.6H₂O in a ratio of 10:1:1 at 37 °C. Aliquots of 100 μ L of samples 10-fold diluted with ultrapure water were added to 3 mL of FRAP reagent in a test tube. After 15 min of incubation at 37 °C, the absorbance was measured at 593 nm in a spectrophotometer (Shimadzu UV 1800, Kyoto, Japan). The results were expressed as mmol of ferrous sulfate equivalent (FSE) per 100 g cherry tomatoes fresh weight (fw).

2.4. Effects on physicochemical parameters

Color was evaluated using a Minolta Chroma Meter CR 200b (Minolta Corp. Tokyo, Japan). Five fruits were used from each sample, with twenty measurements taken from each fruit. The following CIE L* a* b* variables were obtained: L* (lightness), a* (redness–greenness), b* (blueness–yellowness).

The textural parameters were performed on non-irradiated and irradiated samples of five cherry tomatoes (6 measures/sample) using a texturometer (TA-Hdi Stable Micro System, Godalming, UK) equipped with a puncture probe of 2 mm diameter, 1 mm/s speed and a load cell of 25 kg. Results were expressed in Newton (N).

Total Soluble Solids (TSS), pH and titratable acidity were determined for each sample using the extracted juice of five fruits. TSS measurements (12 measurements/sample) were performed with a hand-held refractometer ATAGO (Atago Co, Ltd., Tokyo, Japan) and the results were expressed as "Brix. The pH was measured with a potentiometer Crison-micro pH 2002 (Crison Instruments SA, Barcelona, Spain) using a glass electrode (three measures/sample). The titratable acidity (three measures/sample) was determined by titration and expressed in grams citric acid per 100 g cherry tomatoes fresh weight (fw). The maturity index has been calculated as the ratio TSS to titratable acidity.

2.5. Sensory analysis

An untrained test panel consisting of ten randomly selected individuals (20 < age < 60; 30% smokers, 100% healthy subjects) was performed to assess the sensory quality of samples and factors determining refusal or acceptability of the product by the consumer. Evaluated parameters were: 1) color, 2) odor, 3) flavor, 4) texture, 5) fracturability, 6) firmness, 7) sweetness, 8) acidity, 9) overall output and 10) purchase intention. A hedonic 5 point scale was used, ranging from 1 (dislike extremely) to 5 (like extremely).

2.6. Storage study

In order to evaluate shelf-life of cherry tomatoes, microbiological and physicochemical parameters were assessed immediately after irradiation (0 days of storage; T0), after 7 days (T7) and 14 days (T14) of storage at $4\,^{\circ}$ C as described previously.

2.7. Data analysis

Origin software version 7.5 (OriginLab Corporation, Northampton, USA) was used for data analysis. Data were subjected to analysis of variance (ANOVA) and significant differences among the means were determined by Fisher Post hoc test at a p < 0.05 significant level. All results were expressed as the mean \pm standard deviation. D10-values in kGy, which is the irradiating dose required to reduce microorganisms by 90%, were calculated from the linear regression model of the log of the surviving fractions.

3. Results and discussion

3.1. Microbial inactivation studies

Contamination of tomatoes with microorganisms can occur during fruit development and harvesting, in other words, during pre-harvest and post-harvest stages (Das et al., 2006). Fig. 1 shows the total mesophilic counts for non-irradiated and irradiated cherry tomatoes during storage time. The estimated average bioburden value for the mesophilic microbial population was 2.6 \pm 0.1 log CFU/g. The filamentous fungi were not detected in the analyzed cherry tomato samples. Concerning the stratification assay of bioburden, the microbial growth was only detected in the skin of cherry tomatoes. Other authors reported that populations of Salmonella on dip inoculated tomatoes are largest in the stem scar tissue (Zhuang, Beuchat, & Angulo, 1995), and this microorganism may attach and remain viable during fruit development, thus serving as routes or reservoirs for contaminating ripened fruit (Guo, Chen, Brackett, & Beuchat, 2001). To the best of our knowledge there is no report on the microbial load of cherry tomatoes, however previous studies have cited a bioburden of 4.4 \pm 0.24 log CFU/g for diced Roma tomatoes (Prakash, Manley, DeCosta, Caporaso, & Foley, 2002). This difference may be related to variation in the tomatoes due to season and maturity (Prakash et al., 2002), fruit variety, cultivation, harvest and post-harvest processes.

The mesophilic microbial population of cherry tomatoes showed a linear inactivation kinetics immediately after irradiation (T0; Fig. 1),

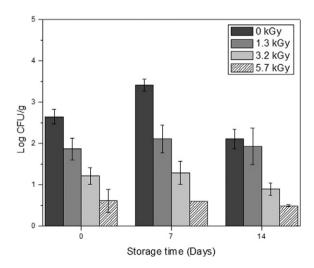


Fig. 1. Total mesophilic counts for non-irradiated and irradiated cherry tomatoes during storage time. Standard deviation bars correspond to 95% confidence intervals about mean values (n=18; $\alpha=0.05$).

with a maximum decimal reduction of 2.2 log CFU/g after irradiation at 5.7 kGy (inactivation efficiency of 99.8%). Similar initial microbial log-reduction results on diced tomatoes were observed by (Prakash et al., 2002), however these authors have cited a dramatic increase in the microbiota of irradiated tomatoes between days 12 and 15 of storage. On the other hand, our data indicated that the observed reduction in the microbial load of cherry tomatoes remained almost constant during the 14 days of storage (Fig. 1), highlighting the potential use of gamma radiation as a post-harvest decontamination treatment of cherry tomatoes.

The microbiota from non-irradiated and irradiated fruits was phenotyped in order to evaluate the dynamics of the microbial communities and its patterns with radiation doses (Table 1). The cherry tomatoes presented an initial microbiota of gram-negative, oxidasenegative, rods (75%) and yeasts (17%). The surviving microbiota of irradiated cherry tomatoes does not seem to be homogenous along storage time, shifting between the prevalence of yeast and Gram-positive cocci to Gram-negative and Gram-positive rods. Similar microbial patterns were observed for the microbiota of lycium fruit after irradiation at different doses (4 up to 14 kGy) of gamma radiation, indicating a significant change on the microbial profiles as the radiation dose increased (Wen, Chung, Chou, Lin, & Hsieh, 2006).

Challenging tests were performed to evaluate the disinfection potential of gamma radiation by simulating conditions of food contamination by microbial pathogens, which have been associated to tomatorelated outbreaks (Canadian Food Inspection Agency, 2012; Center for Science in the Public Interest, 2010; Solano et al., 2013; Valadez et al., 2012). The inoculated bacterial strains followed a linear inactivation kinetics by gamma radiation on cherry tomatoes, which allowed to estimate D10-values of 0.30 \pm 0.01 kGy for Salmonella Typhimurium, 0.45 ± 0.02 kGy for Staphylococcus aureus and 0.71 ± 0.04 kGy for Escherichia coli. Furthermore, the obtained results after irradiation at 3.2 kGy, indicated a decrease of 5, 7 and 11 log CFU/g on the populations of E. coli, S. aureus and S. Typhimurium inoculated on cherry tomatoes, respectively. The effects of non-thermal treatments on microbial viability on the inoculated cherry tomatoes have been cited (Brilhante São José & Dantas Vanetti, 2012; Song et al., 2011). Namely, the populations of S. Typhimurium and E. coli O157:H7 in inoculated cherry tomatoes were reduced by approximately 2.3 log CFU/g after treatment with aqueous ClO₂, and by 2.6 log CFU/g, after treatment with UV-C compared to the control (Song et al., 2011). Other authors (Brilhante São José & Dantas Vanetti, 2012) mentioned that the combined treatment of ultrasound and 40 mg/L peracetic acid resulted in the highest reduction of the natural contaminant population and a reduction of adherent Salmonella Typhimurium ATCC 14028 by 3.9 log CFU/g. Nevertheless, in this study the obtained log-reductions on microbial viability highlighted the efficacy of gamma radiation on the inactivation of potential pathogenic microorganisms. Although, the radiation resistance of microorganisms can differ from species to species and between strains of the same species, depending on both biotic and abiotic factors (Cabo Verde et al., 2010). The obtained D10-values for the inoculated bacteria were in the range of the ones cited in literature, with exception of *E. coli* that demonstrated in cherry tomatoes a higher radioresistance (Chirinos, Vizeu, Destro, Franco, & Landgraf, 2002; Lamb, Gogley, Thompson, Solis, & Sen, 2002; Prakash, Johnson & Foley, 2007; Rajkowski & Thayer, 2000). These radiation sensitivities differences among microorganisms are correlated to their inherent diversity with respect to the chemical and physical structure as well as their capacity to recover from radiation injuries (Cabo Verde et al., 2010).

3.2. Total phenolic content and antioxidant activity

The obtained results of Total Phenolic content (TP) and antioxidant activity of cherry tomatoes before and after irradiation and during storage time are presented in Table 2.

Phenolic compounds are important because they contribute to the nutritional and sensory quality of fruits (Shahbaz et al., 2014). The obtained TP value for the non-irradiated cherry tomatoes was 121.5 mg GAE/100 g of FW, and no significant trend was verified along the 14 days of refrigerated storage. Previous studies on postharvest ripening of cherry tomatoes also indicated that total phenolics remained stable during storage at refrigerated temperatures during 3 weeks (Slimestad & Verheul, 2005). Regarding the effect of irradiation doses on phenolic content of cherry tomatoes, it was verified that the highest and the lowest TP values were obtained for the irradiated samples at 3.2 kGy (T0) and 5.7 kGy (T7), respectively. Although, only the phenolic content corresponding to cherry tomatoes irradiated at 3.2 kGy (T0) was significantly (p < 0.05) higher than the TP values determined for the other irradiated and non-irradiated samples (exception for cherry tomatoes after irradiation (T0) at 1.3 kGy). According to Schindler, Solar, and Sontag (2005) gamma irradiation reduced the concentration of the phenolic compounds in traditional tomato varieties; however, this change was smaller than the naturally occurring differences. Diverse results have been published for the irradiation effect on phenolic compounds in foods. Some authors mentioned a significant decrease in the total phenols of fruit juice immediately after irradiation at 5 kGy (Shahbaz et al., 2014; Song et al., 2006), as can be observed in Fig. 2. In contrast, El-Samahy, Youssef, Askar, and Swailam (2000) reported that the concentration of the total phenolic compounds was higher in irradiated mangoes (0.5-1.5 kGy) compared to the

 Table 1

 Frequency of the morphological phenotypes of the isolates from non-irradiated and irradiated cherry tomatoes with storage time.

Storage time (days)	Phenotype	% of total microbiota Dose (kGy)				
		0	1.3	3.2	5.7	
0	Gram positive, catalase positive cocci	5.0	4.9	24.6	58.5	
	Gram negative, catalase positive cocci	0.8	1.2	3.7	nd	
	Gram positive rods	1.3	3.7	3.7	nd	
	Gram negative, oxidase negative rods	75.2	13.7	2.3	41.5	
	Yeasts	17.4	76.5	65.7	nd	
7	Gram positive, catalase positive cocci	2.4	nd	nd	55.6	
	Gram negative, catalase positive cocci	nd	0.8	nd	nd	
	Gram positive rods	nd	0.8	0.1	nd	
	Gram negative, oxidase negative rods	62.3	nd	nd	44.4	
	Yeasts	35.3	98.4	99.9	nd	
14	Gram positive, catalase positive cocci	6.1	1.7	20.0	nd	
	Gram negative, catalase positive cocci	nd	nd	nd	nd	
	Gram positive rods	18.4	5.2	35.0	50.0	
	Gram negative, oxidase negative rods	12.2	79.3	10.00	50.0	
	Yeasts	63.3	13.8	35.0	nd	

Table 2Total phenolic content and antioxidant activity measured by FRAP and DPPH, in extracts of non-irradiated and irradiated cherry tomatoes during storage time.

Storage time (days)	Dose (kGy)	Total phenolic content	Antioxidant activity		
		mg GAE/100 g fw	mmol FSE/100 g fw	EC50	
0	0	121.5 ± 3.7 ^{b,c}	1.50 ± 0.04 ^{b,d,e}	0.11 ± 0.03^{a}	
	1.3	$137.9 \pm 5.4^{a,c}$	$1.54 \pm 0.02^{b,c,d}$	0.14 ± 0.01^a	
	3.2	143.9 ± 7.2^{a}	$1.67 \pm 0.05^{a,c,d}$	0.14 ± 0.02^{a}	
	5.7	$116.5 \pm 0.4^{b,c}$	$1.39 \pm 0.02^{\mathrm{d,f}}$	0.18 ± 0.05^{a}	
7	0	$106.8 \pm 3.0^{b,d}$	$1.56 \pm 0.02^{a,b}$	0.16 ± 0.09^{a}	
	1.3	$109.5 \pm 1.7^{\mathrm{b,d}}$	1.69 ± 0.04^{a}	0.18 ± 0.02^{a}	
	3.2	$104.1 \pm 0.5^{\mathrm{b,d}}$	$1.64 \pm 0.03^{a,b}$	0.18 ± 0.04^{a}	
	5.7	90.9 ± 7.9^{d}	$1.49 \pm 0.02^{\mathrm{b,d}}$	0.21 ± 0.11^{a}	
14	0	115.0 ± 2.5^{b}	$1.51 \pm 0.01^{\mathrm{b,d}}$	0.23 ± 0.13^{a}	
	1.3	$113.0 \pm 4.4^{b,d}$	$1.53 \pm 0.03^{\mathrm{b,c,f}}$	0.20 ± 0.12^{a}	
	3.2	$119.1 \pm 4.2^{b,c}$	1.60 ± 0.04^{a}	0.18 ± 0.07^{a}	
	5.7	$103.6 \pm 4.0^{b,d}$	$1.35 \pm 0.01^{d,e}$	0.21 ± 0.10^a	

Values within columns sharing a common superscript letter are not significantly different (p>0.05). Listed values are averages of triplicate analyses (n=3) of a single extract \pm SD.

control. As mentioned elsewhere, the increase effect of gamma radiation on phenolic content after irradiation could be explained by a structural alteration due to an immediate oxidation of the phenolic compounds that can play an antioxidant role by reducing the free radicals and the reactive oxygen species induced by irradiation (Song et al., 2006). At higher radiation doses, the apparent decrease of TP content may be due to a slight degradation effect of gamma radiation.

As for phenolic content, no significant change in the amount of anti-oxidant activity (methanol soluble antioxidants) was found during storage for non-irradiated samples (Table 2). According to Slimestad and Verheul (2005) no effect of post-harvest ripening was verified on the content of methanol soluble antioxidants of the cherry tomato fruits. With regard to the effect of irradiation on the antioxidant activity, the irradiated samples at 1.3 kGy after 7 days of storage presented the highest value in terms of FRAP assay, that was significantly different from the control sample of non-irradiated cherry tomatoes (0 kGy; T = 0), but not from non-irradiated samples stored for 7 days (0 kGy; T = 7). The lowest antioxidant activity by FRAP assay was obtained for the irradiated samples at 5.7 kGy, however there was no significant differences between these values and the ones measured for the non-irradiated samples. Besides that, DPPH results expressed by EC₅₀

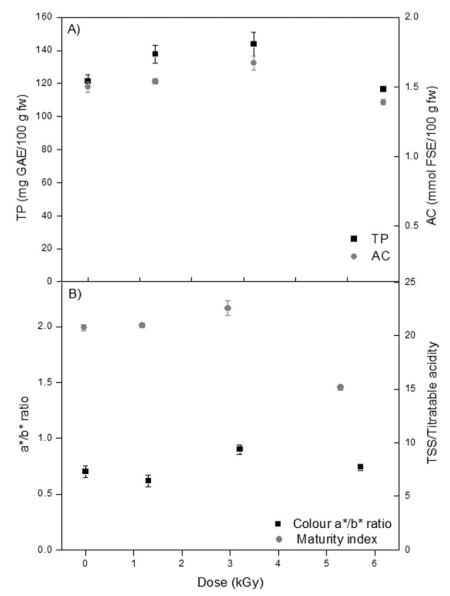


Fig. 2. Trend of some physicochemical attributes of cherry tomatoes after irradiation (T0): A) Total phenolic content (TP) and antioxidant activity (AC) by FRAP assay; and B) colour by a^*/b^* ratio and firmness. Standard deviation bars correspond to 95% confidence intervals about mean values (3 < n < 12; $\alpha = 0.05$).

indicated no significant difference (p > 0.05) among the antioxidant activity of non-irradiated and irradiated cherry tomatoes throughout the 14 days of storage time. To the best of our knowledge, this is the first report on the effects of gamma radiation on the antioxidant activity of cherry tomatoes. Most of the natural antioxidants are multifunctional; therefore, for a more reliable evaluation, it is important to perform different antioxidant activity assessments to give proper consideration to the various mechanisms of antioxidant action. For example, FRAP does not measure compounds that act by radical quenching (H transfer) like thiol antioxidants; TP includes nonphenolic compounds; and DPPH interferes with carotenoids at 513 nm (Balogh, Hegedűs, & Stefanovits-Bányai, 2010). The obtained results could be related with TP content (Fig. 2) and can suggest that the phenolic compounds are the main responsible for the FRAP activity. As mentioned by other authors (Csambalik et al., 2014) the existence of stress factors, in which irradiation can be included, can induce the production of polyphenols leading to an increase of TP and antioxidant activity. On the other hand, Toor and Savage (2006) reported that storage at refrigerated temperatures (7 °C) inhibited the accumulation of lycopene in tomatoes, fact that could explain the decreasing trend of TP and antioxidant activity observed during storage at 4 °C. Nevertheless, the antioxidant capacity of tomatoes depends on a large number of phytochemical compounds and the interactions that occur between them (Odriozola-Serrano, Soliva-Fortuny & Martín-Belloso, 2008) resulting from numerous factors such as variety, growing and environmental conditions (Gonzalez-Cebrino et al., 2011).

3.3. Evaluation of physicochemical parameters

3.3.1. Color and texture assessments

Surface color is one of the most appealing factors that influence the consumer in the purchase of fresh food. Therefore, the evaluation of the global color properties was done for irradiated and non-irradiated cherry tomatoes and results are shown in Table 3. Considering the a* parameter, no significantly (p < 0.05) differences were detected with irradiation comparatively with the control (0 kGy; T=0). Although, the a^* value from non-stored irradiated cherry tomatoes at 1.3 kGy differ significantly (p > 0.05) from the a* values of the other non-stored irradiated samples (3.2 kGy and 5.7 kGy; T = 0). After irradiation (T0), was observed a significant decrease (p > 0.05) on the b* value of 1.3 kGy irradiated cherry tomatoes comparatively to control cherry tomatoes (0 kGy; T = 0). The decrease of b* values in tomatoes is assumed to reflect the biosynthesis of lycopene and after a certain point is correlated with progression of the ripening (Liu, Zabaras, Bennett, Aguas, & Woonton, 2009). For both chromaticity coordinates it was verified a decreasing tendency with storage time, that was significantly (p > 0.05) different for the b^* parameter. This result was expected because color is strongly influenced by fruit ripeness. Also, this can indicate that irradiation at the applied gamma radiation doses can retain the red color of cherry tomatoes and did not delay their storage ripening process. In the case of tomato ripening, different colors are present simultaneously since chlorophyll is degraded from green to colorless compounds at the same time that carotenoids are synthesized from colorless precursor (phytoene) to carotene (pale yellow), lycopene (red), β -carotene (orange), and xanthophylls and hydroxylated carotenoids (yellow) (Giuliano, Bartley, & Scolnik, 1993). No significant differences were observed in lightness (L*) values between non-irradiated and irradiated cherry tomatoes, either immediately after irradiation or after/during storage.

The ratio a*/b* is a good indicator of color in tomatoes, expressing well the changes in color that occur (Akter & Khan, 2012). After irradiation it was verified an increase of a*/b* ratio values (Fig. 2), which with storage time became significantly different (p > 0.05) for the applied gamma radiation doses of 3.2 kGy and 5.7 kGy. According to the literature (Akter & Khan, 2012), ripe traditional tomatoes presented values for a*/b* of 1.19, and a minimum value of 0.92 was set for fruit in a stage of commercial ripeness. A correlation has been established between lycopene content and the color index a*/b*, such that those tomato varieties with higher color indices were also those with greater lycopene levels (Akter & Khan, 2012; Misra, Keener, Bourke, Mosnier, & Cullen, 2014). Regarding the obtained results the significant increase of tomato color index with irradiation and storage indicated the possibility of higher lycopene levels, which needs further investigation.

The mean values of peak force (N) required for puncturing (break) the fruits is presented in Table 3. The firmness of irradiated group of produce was lower than that of control non-irradiated tomatoes (Fig. 2) and this difference was significant (p < 0.05) for the 5.7 kGy irradiated cherry tomatoes. An insignificant difference (p > 0.05)between the firmness values of cherry tomatoes was recorded during storage period, meaning that the tissue structure of the produce remained intact. However, after 14 days of storage the irradiated cherry tomatoes (all doses) presented significantly lower firmness values than control samples (0 kGy, non-stored and stored). Previous studies on traditional tomatoes, also reported firmness loss of the irradiated samples compared with their non-irradiated counterparts (Akter & Khan, 2012; Magee, Caporaso, & Prakash, 2003). In contrast, other authors indicated no significant differences ($p \le 0.05$) in flesh firmness among the irradiated tomato fruits at 24 days of storage (Adam, Elbashir, & Ahmed, 2014). Moreover, the loss of firmness of cherry tomatoes with plasma treatments was also reported (Misra et al., 2014).

3.3.2. Total soluble solids, titratable acidity and pH assessments

The amount of Total Soluble Solids (TSS) in the fruit (measured in Brix units) is a parameter of great agronomic importance in tomato. Cherry tomatoes are small and tasty tomatoes and the taste advantage is primarily connected to their high content of soluble solids (sugars)

Table 3CIE L* a* b* parameters and colour index of the skins, and texture parameter of non-irradiated and irradiated cherry tomatoes during storage time. Mean values ± SD (standard deviation) are presented.

Storage time (days)	Dose (kGy)	Colour parameters	Firmness			
		L* (lightness)	a* (redness-greenness)	b* (blueness-yellowness)	a*/b*	Force (N)
0	0	$37.97 \pm 0.59^{a,b}$	$16.43 \pm 0.82^{a,b,c,d}$	$24.43 \pm 0.82^{a,b}$	0.70 ± 0.05°	$5.33 \pm 0.19^{a,b,c}$
	1.3	$36.71 \pm 0.35^{b,c}$	13.57 ± 1.09^{d}	$22.02 \pm 0.41^{c,d}$	0.62 ± 0.05^{c}	$4.40 \pm 0.28^{c,e}$
	3.2	35.95 ± 0.17^{c}	$19.97 \pm 0.95^{a,b,c}$	$22.27 \pm 0.36^{b,c}$	$0.90 \pm 0.04^{a,b}$	$4.42 \pm 0.17^{c,d}$
	5.7	37.73 ± 0.39^{b}	$18.27 \pm 0.84^{a,b,c}$	24.74 ± 0.63^{a}	0.74 ± 0.03^{c}	3.95 ± 0.18^{d}
7	0	39.37 ± 0.35^{a}	$14.12 \pm 0.82^{c,d}$	$20.32 \pm 0.46^{c,e}$	0.71 ± 0.05^{c}	6.04 ± 0.17^{a}
	1.3	$38.01 \pm 0.34^{a,b}$	$14.10 \pm 0.71^{c,d}$	$17.83 \pm 0.46^{f,g}$	0.80 ± 0.04^{c}	$5.07 \pm 0.15^{\mathrm{b,c}}$
	3.2	37.63 ± 0.24^{b}	$15.60 \pm 0.64^{\mathrm{b,c,d}}$	$19.04 \pm 0.32^{c,f,g}$	$0.83 \pm 0.04^{a,b}$	$4.87 \pm 0.23^{c,d}$
	5.7	37.76 ± 0.22^{b}	$17.21 \pm 0.61^{b,c}$	$18.92 \pm 0.49^{c,f}$	$0.91 \pm 0.03^{a,b}$	3.12 ± 0.20^{e}
14	0	39.26 ± 0.43^{a}	$15.52 \pm 0.92^{\mathrm{b,c,d}}$	$20.15 \pm 0.58^{c,d,f}$	$0.79 \pm 0.05^{\mathrm{b,c}}$	$5.91 \pm 0.36^{a,b}$
	1.3	$37.89 \pm 0.33^{a,b}$	13.54 ± 0.57^{d}	$18.33 \pm 0.56^{e,f,g}$	0.75 ± 0.04^{c}	4.08 ± 0.07^{e}
	3.2	37.58 ± 0.21^{b}	18.19 ± 0.48^{b}	$18.89 \pm 0.44^{ m e,f,g}$	$0.97 \pm 0.03^{a,b}$	$4.02 \pm 0.13^{d,e}$
	5.7	$36.57 \pm 0.29^{b,c}$	$16.83 \pm 0.35^{\mathrm{b,c,d}}$	16.86 ± 0.25^{g}	1.00 ± 0.02^{a}	3.28 ± 0.13^{e}

For each parameter (columns) the values between treatments that have the same letters are not considered significantly different (p > 0.05).

and titrable acidity (Slimestad & Verheul, 2005). The average values of TSS, pH and titratable acidity for irradiated and non-irradiated cherry tomatoes analyzed immediately after irradiation, and after storage at 4 °C are shown in Table 4.

The TSS content of cherry tomatoes was found to be 7.6 °Brix, which is in accordance with the literature (Slimestad & Verheul, 2005). Moreover, it was observed an increasing tendency of TSS values for the radiation doses of 3.2 kGy and 5.7 kGy, although only significantly different (p < 0.05) for the highest dose applied. This may be related to the radiolytic effect of irradiation on cherry tomatoes sugars, leading to an increase of % of sugar. Total soluble solids are predominantly influenced by the amount of sugars in the fruits (Adam et al., 2014). Nevertheless, the storage time seems to have no influence on TSS values except for the highest irradiation dose (Table 4). As mentioned by other authors, radiation did not affect TSS (% sugar) in stored traditional tomatoes (Akter & Khan, 2012).

The pH of the cherry tomatoes was found to decrease significantly (p < 0.05) with irradiation at the highest dose. However, the obtained pH values are comparable to those reported in literature (Misra et al., 2014; Rodriguez-Lafuente, Nerin, & Batlle, 2010). Storage time seems to have no significant (p > 0.05) effect on tomatoes pH, except for the samples irradiated at 3.2 kGy and 5.7 kGy, for which it was verified a significant (p < 0.05) increase. An increase in pH of cherry tomatoes in storage under natural conditions has also been reported by Rodriguez-Lafuente et al. (2010). The change in pH could be attributed to the metabolic changes and water loss in the tomatoes (Garcia, Casariego, Diaz, & Roblejo, 2014). A significant effect of irradiation and storage time on titratable acidity was only verified for cherry tomatoes irradiated at 5.7 kGy, except for the samples stored for 7 days. The maturity index (ratio TSS to titratable acidity) gives a good indication of tomatoes ripeness. The cherry tomatoes irradiated at 3.2 kGy presented the higher maturity indexes intra-storage time, and stood out in this study as significantly different (p < 0.05) after 14 days of storage. To the best of our knowledge there is no literature regarding irradiation of cherry tomatoes, however the obtained results are in agreement with previous studies performed in irradiated traditional tomatoes (Akter & Khan, 2012).

3.4. Sensorial analysis

The characterization of the sensory properties of non-irradiated and irradiated cherry tomatoes is presented in a radar chart constructed with the ratings obtained for the different parameters evaluated by the non-trained panelists (Fig. 3).

Cherry tomatoes irradiated at 5.7 kGy showed the lowest score almost for all evaluated parameters (color was the exception), corroborating with the results obtained from quality parameters measurements. Generally, non-irradiated and 3.2 kGy irradiated cherry tomatoes were almost equally rated, which includes the purchase

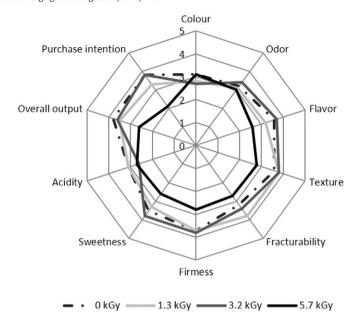


Fig. 3. Sensory evaluation for non-irradiated and irradiated cherry tomatoes immediately after irradiation (T0). Panel members (10 untrained panellists) were asked to evaluate exterior and interior colour, odor, flavor, texture, fracturability, firmness, sweetness, acidity, overall output and purchase intention. A hedonic scale was used, ranging from 1 (dislike extremely) to 5 (like extremely).

intention. The samples irradiated at 3.2 kGy were rated as sweeter than non-irradiated, which is in agreement with the obtained Brix values and Maturity index.

4. Conclusion

In summary, the obtained data for cherry tomatoes indicated that an irradiation dose of 3.2 kGy did result in a major impact on the benefit of reducing microbiota by 2 log (99% inactivation) and a potential decrease of 5–11 log of foodborne pathogens load with a minor effect on the fruit sensory and quality attributes. The implementation of this emergent post-harvest process for cherry tomatoes could represent a shelf-life extension up to 14 days at refrigerated conditions. A possible added-value of enhancing cherry tomatoes lycopene level with irradiation was hypothesized, but needs further studies.

Due to health benefits of rich antioxidant fruits the applicability of irradiation technology as post-harvest food safety processing technology may serve as a step forward to increase the variety, availability and acceptability of foods for immunocompromised patients and other target groups with special dietary needs.

 Table 4

 Average values of total soluble solids (TSS), pH and titratable acidity of non-irradiated and irradiated cherry tomatoes during storage time. Mean values ± SD (standard deviation) are presented.

Storage time (days)	Dose (kGy)	TSS (°Brix)	pH	Titratable acidity (g citric acid/100 g-fw)	Maturity index
0	0	7.6 ± 0.1 ^{b,d}	4.610 ± 0.015 ^{c,d}	$3.640 \pm 0.001^{c,d,e}$	$20.8 \pm 0.3^{b,c,d,e,f}$
	1.3	6.7 ± 0.1^{d}	$4.630 \pm 0.061^{c,d}$	$3.220 \pm 0.140^{\mathrm{d,e}}$	$20.9 \pm 0.2^{b,c,d,e,f}$
	3.2	$7.3 \pm 0.2^{\rm d}$	4.567 ± 0.003^{d}	$3.220 \pm 0.140^{\mathrm{d,e}}$	$22.6 \pm 0.7^{b,c,d,e}$
	5.7	9.8 ± 0.2^{a}	$4.217 \pm 0.003^{\mathrm{f}}$	6.440 ± 0.001^{a}	$15.2 \pm 0.3^{\rm f}$
7	0	7.1 ± 0.2^{d}	$4.640 \pm 0.006^{\mathrm{b,c,d}}$	$3.640 \pm 0.280^{c,d,e}$	$19.5 \pm 0.5^{b,d,f}$
	1.3	6.7 ± 0.3^{d}	$4.687 \pm 0.018^{\mathrm{b,c}}$	$3.920 \pm 0.001^{c,d}$	17.1 ± 0.8^{f}
	3.2	$8.8 \pm 0.3^{a,b}$	4.727 ± 0.012^{b}	$3.500 \pm 0.140^{ m d,e}$	$25.2 \pm 0.7^{b,c,e}$
	5.7	$8.8 \pm 0.3^{a,b,c}$	4.313 ± 0.003^{f}	$4.060 \pm 0.140^{\circ}$	$21.6 \pm 0.6^{b,c,d}$
14	0	$7.7 \pm 0.2^{c,d}$	$4.710 \pm 0.012^{b,c}$	3.360 ± 0.001^{d}	$22.8 \pm 0.5^{\mathrm{b,c}}$
	1.3	$7.4 \pm 0.2^{ m d}$	4.563 ± 0.009^{d}	$3.220 \pm 0.001^{\mathrm{d,e}}$	$22.9 \pm 0.7^{\rm b}$
	3.2	$7.5\pm0.4^{ m d}$	4.973 ± 0.003^{a}	$2.240 \pm 0.001^{\mathrm{f}}$	33.6 ± 1.9^{a}
	5.7	$8.8 \pm 0.2^{a,b,c}$	4.450 ± 0.006^{e}	$4.760 \pm 0.001^{\mathrm{b}}$	$18.4 \pm 0.5^{\rm d,f}$

The values between treatments that have the same letters are not considered significantly different (p > 0.05).

Acknowledgments

This work was developed within the Coordinated Research Project D6-RC-1163.2 financed by the International Atomic Energy Agency (IAEA). It was also supported by Fundação para a Ciência e a Tecnologia through financial support of RECI/AAG-TEC/0400/2012 and UID/Multi/04349/2013 projects.

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