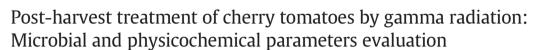
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ABSTRACT

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. Therefore, these results suggest that the irradiation articles 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.

* Corresponding author. *E-mail address:* sandracv@ctn.tecnico.ulisboa.pt (S. Cabo Verde). 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 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 °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

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