



Effects of gamma radiation, individually and in combination with bioactive agents, on microbiological and physicochemical properties of ground beef

Samia Ayari ^{a, b}, Jaejoon Han ^c, Khanh Dang Vu ^a, Monique Lacroix ^{a, *}

^a Research Laboratory of Sciences Applied to Food, INRS-Institut Armand Frappier, Canadian Irradiation Centre, 531 Boulevard des Prairies, Laval, Quebec H7V 1B7, Canada

^b National Centre for Nuclear Sciences and Technologies (CNSTN), Tunis Cedex 2020, Tunisia

^c Department of Food Bioscience and Technology, College of Life Sciences and Biotechnology, Korea University, Seoul 136-701, Republic of Korea

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ABSTRACT

Effects of gamma irradiation (2 kGy) on microbiological and physicochemical properties of ground beef were investigated individually or in combination with bioactive compounds consisting of cinnamaldehyde, ascorbic acid, and sodium pyrophosphate decahydrate. Meat samples were mixed with cinnamaldehyde (1.47%, w/w), cinnamaldehyde plus ascorbic acid (0.5%, w/w), or cinnamaldehyde plus sodium pyrophosphate decahydrate (0.1%, w/w), followed by irradiation at a dose of 2 kGy. Microbiological and physicochemical properties of samples were analyzed. The results demonstrated that combined treatments using irradiation and bioactive compounds could significantly decrease the microbial load of meat samples compared with that of the untreated control samples. Moreover, the combined treatments did not cause any significant changes in physical or chemical properties of meat samples. Irradiation, both with and without cinnamaldehyde treatment, caused significant increases in concentrations of thiobarbituric reactive substances and peroxide in treated samples, compared with the concentrations of these substances in the control samples; however, in the presence of ascorbic acid, the pro-oxidative effect of irradiation treatment on ground beef was overcome.

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1. Introduction

Meat and meat products are particularly susceptible to contamination with food pathogens due to their high nutritional value. During production, processing, distribution, and storage, food undergoes deterioration through chemical and microbial processes (Sanchez-Escalante, Djenane, Torrescano, Beltran, & Roncales, 2001). Gamma irradiation has been used successfully for controlling microbial contamination in meat and meat products (Konteles, Sinanoglou, Batrinou, & Sflomos, 2009; Lee, Jo, Shin, Kim, & Byun, 2006). For example, Fu, Sebranek, and Murano (1995a) found that medium-dose irradiation (1.8–2.0 kGy) accompanied by storage at 7 °C destroyed *Yersinia enterocolitica* and *Escherichia coli* O157:H7 in beef steaks and ground beef. Fu, Sebranek, and

Murano (1995b) also found that in cooked pork chops and cured hams, low-dose irradiation (0.75–0.90 kGy) reduced the number of viable cells of *Listeria monocytogenes* by more than 2 log and those of *Salmonella typhimurium* by 1–3 log. In the same study, after a medium-dose irradiation (1.8–2.0 kGy), pathogen populations were reduced to undetectable levels. Moreover, the authors demonstrated that meat quality attributes (color and odor) were not affected by irradiation (Fu et al., 1995b). Konteles et al. (2009) evaluated the antilisterial effects of gamma irradiation on Feta cheese; the authors conducted investigations of cheese samples contaminated with *L. monocytogenes* (10³ CFU/ml), which were subjected to vacuum-packaging, exposed to irradiation doses of 1.0, 2.5, and 4.7 kGy, and stored at 4 °C for 1 month. The authors found that 2.5 kGy and 4.7 kGy doses of irradiation reduced *L. monocytogenes* counts to below the detection limit and had no effect on the texture of the Feta cheese.

The deterioration of meat products by chemical reaction is mainly due to oxidation resulting in degradation of fats in raw meat via the peroxidation of unsaturated fatty acids originating from

List of abbreviations: EO, essential oil; TVBN, total volatile basic nitrogen; PV, peroxide value.

* Corresponding author.

E-mail address: monique.lacroix@iaf.inrs.ca (M. Lacroix).

phospholipids (Sanchez-Escalante et al., 2001). Moreover, oxymyoglobin can be oxidized to metmyoglobin during storage or by free radicals (such as $\cdot\text{OH}$), which cause meat products to turn brown and appear unattractive (Giroux et al., 2001; Sanchez-Escalante et al., 2001). However, the rate and level of oxidative deterioration can be reduced by treatment with antioxidants (Djenane, Sánchez-Escalante, Beltrán, & Roncalés, 2003; Giroux et al., 2001), which protect lipids from oxidation and stabilize oxymyoglobin (Sanchez-Escalante et al., 2001).

Because of widespread public perception that natural products are safer for consumption than artificial additives, the use of natural antioxidants to limit the chemical deterioration of meat products has been increasing (Ahn, Grün, & Mustapha, 2007). Natural antioxidants can be added directly to meat or meat products during processing. Ascorbic acid possesses antioxidant properties (i.e., it can scavenge free radicals) and can therefore be used in meat products to preserve their initial appealing color (Djenane et al., 2003; Sanchez-Escalante et al., 2001). Giroux et al. (2001) evaluated the effects of ascorbic acid at different concentrations (0.03–0.5%, w/w) and with different doses of irradiation (0.5–4 kGy) on microbial growth, color coordinates, and sensory characteristics of beef patties during storage at 4 °C. The authors found significant reductions in the numbers of aerobic plate counts and total coliforms. The authors found that incorporation of ascorbic acid into the meat before irradiation resulted in a significant stabilization of color parameters, while irradiation treatment had detrimental effects on the redness, yellowness, and hue angle values of meat (Giroux et al., 2001). Sodium pyrophosphate in combination with thermal inactivation has been reported to be an effective antibacterial agent against *L. monocytogenes* in pork slurry and ground pork (Lihono, Mendonca, Dickson, & Dixon, 2001) and against *Clostridium perfringens* (Akhtar, Paredes-Sabja, & Sarker, 2008).

Recent developments in food irradiation technology have included the use of combined treatments to reduce irradiation doses required to eliminate pathogenic bacteria and/or reduce microbial loads (Ayari, Dussault, & Hayouni et al., 2012; Lacroix & Ouattara, 2000). Combining irradiation with various natural or synthetic active compounds can increase the radiosensitization of food pathogens (Lacroix, Ouattara, Saucier, Giroux, & Smoragiewicz, 2004). Many natural extracts such as essential oils (EOs) from edible and medicinal plants, herbs, and spices possess antimicrobial properties and could serve as sources for antimicrobial agents for use against food spoilage and pathogens (Bagamboula, Uyttendaele, & Debevere, 2003). The hydrophobicity of EOs enables them to partition lipids of the cell membrane and mitochondria, rendering them permeable and causing leakage of cell contents (Burt, 2004). In fact, it has been demonstrated that the combination of several EOs with irradiation could increase the shelf life and reduce the pathogenic contamination of food products (Lacroix et al., 2004; Ouattara, Sabato, & Lacroix, 2002). Cinnamaldehyde is the main component of cinnamon EO with a high antimicrobial activity against diverse bacteria, both gram-positive and gram-negative, including *E. coli* O157:H7 (Amalrajou et al., 2010), *Bacillus cereus* (Ayari, Dussault, Jerbi, Hamdi, & Lacroix, 2012; Valero & Francés, 2006), and *Salmonella enterica* (Ravishankar et al., 2010). Our previous study (Ayari, Dussault, & Jerbi et al., 2012) revealed that the irradiation of raw beef meat pre-treated with cinnamaldehyde at 1.47% (w/w), the concentration needed to reduce by 1 log the population of *B. cereus* spores (ATTC 7004) inoculated in minced beef, produced an inhibition of the growth of *B. cereus* count during refrigerated storage. However, there has been a limited amount of research on the use of cinnamaldehyde, especially in combination with antioxidant treatment and gamma irradiation, to reduce microbial loads and preserve

physicochemical properties of meat products.

Therefore, the objective of this study was to evaluate the effects of ionizing radiation individually and in combination with bioactive agents on microbiological and physicochemical properties of ground beef. The bioactive compounds were an antimicrobial agent (cinnamaldehyde), an antioxidant agent (ascorbic acid), and an additive agent for water retention (sodium pyrophosphate decahydrate). The microbial analysis consisted of total mesophilic bacteria, psychrotrophic bacteria, yeast, and molds. The physicochemical properties studied consisted of physical properties (pH, water-holding capacity, and humidity), chemical composition (total protein, fat content, heme iron, total volatile basic nitrogen [TVBN], and free amino acids), lipid oxidation, total acidity, and color of meat samples.

2. Material and methods

2.1. Bioactive agents and sample preparation

Cinnamaldehyde was purchased from Fisher Scientific (Loughborough, Leicestershire, UK). L-ascorbic acid was purchased from MP Biomedicals (Parc d'innovation, Illkirch, France). Sodium pyrophosphate decahydrate was purchased from Acros Organics (Geel, Belgium). Sterility of the active compounds was verified by pouring on tryptic soy agar (Difco Laboratories, Detroit, MI, USA). Fresh ground beef containing 5% fat was purchased at a local grocery store in Tunisia and transported to the National Center for Nuclear Sciences and Technologies, Sidi Thabet, Tunisia, under refrigerated conditions in an ice cooler. Beef samples weighing 100 g each were individually packaged in polyethylene bags. Meat portions were treated with cinnamaldehyde (1.47%, w/w), ascorbic acid (0.5%, w/w), and sodium pyrophosphate decahydrate (0.1%, w/w). Samples were blended for 2 min at medium speed in a Stomacher[®] laboratory blender (LES400, Seward, Worthing, UK) and then irradiated at 2 kGy.

2.2. Irradiation treatment

The aerobically packaged ground beef samples were irradiated at the National Center for Nuclear Sciences and Technologies using a ⁶⁰Co source with a dose rate of 150 Gy/min under unified conditions of temperature (25 ± 1 °C) and humidity (approximately 50%). The samples were placed on ice plaques (1 cm thickness) to avoid an increase in temperature during irradiation. A reference standard Fricke dosimeter system was used for dose distribution measurements. After irradiation, the irradiated and non-irradiated meat samples were immediately stored at 4 ± 1 °C and tested periodically (days 0, 7, 12, and 21) for microbial growth. Biochemical characteristics (lipid oxidation, TVBN, color, etc.) were evaluated immediately after irradiation.

2.3. Microbiological analysis

Each sample of ground beef (10 g) was weighed aseptically and homogenized for 2 min in 90 ml of sterile peptone water (0.1%, w/v, Biokar Diagnostics, Beauvais, Cedex, France) using a Stomacher[®] laboratory blender (LES400, Seward). Serial dilutions of the homogenate were prepared and appropriate dilutions were spread plated onto sterile Petri plates. Colony forming units of total aerobic mesophilic and psychrophilic bacteria were determined by plating dilutions of the ground beef homogenates on plate count agar medium and incubating at 30 °C for 3 days and 7 °C for 7 days, respectively. Total and fecal coliforms were counted on violet red bile lactose agar medium (Biokar Diagnostics) after incubation for 24 h at 30 °C and 44 °C, respectively. Total molds and yeasts were

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