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# Antioxidant activity of carrot juice in gamma irradiated beef sausage during refrigerated and frozen storage

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#### ABSTRACT

The antioxidant activity of carrot juice in gamma irradiated beef sausage was studied. Four batches of beef sausage were prepared, in which the first batch was formulated with water as the control. The other batches were formulated with unconcentrated carrot juice, carrot juice concentrated by 35% and 60%, respectively. Samples were irradiated at doses of 0, 3 and 4.5 kGy. Then the extent of oxidation in raw sausages was determined during refrigerated and frozen storage through the determination of peroxide value (PV) and thiobarbituric acid reactive substances (TBARS) for lipid oxidation and carbonyl content for protein oxidation. The raw sausages were sensory evaluated during storage for colour, appearance and odour, while the grilled samples were sensory evaluated for their colour, taste, texture and juiciness post-treatment only. Irradiation and storage significantly increased the PV, TBARS and carbonyl content in the samples formulated with water. The carrot juice significantly decreased the oxidative processes in the samples proportionally to the juice's concentration. Furthermore, the sausages that were formulated with carrot juice had a high acceptable sensory scores as compared with the control samples.

#### 1. Introduction

Sausage is one of the meat products that is gaining popularity and present in the diet of different cultures because of convenience, variety and economy. It takes little time in preparation, with some sausage being ready to serve, and others needing only to be warmed (Mercadante, Capitani, Decker, & Castro, 2010). However, there is increasing awareness of the risks involving microbiological contamination of meat and meat products which constitute a major source for pathogens that cause food-borne illness to human. Considering a series of recent outbreaks of pathogenic bacteria in meat, the expanded application of irradiation in meat and meat products becomes especially important to improve safety and public confidence (Lim, Seol, Jeon, Jo, & Lee, 2008).

Food irradiation is proven to be the best effective physical decontamination technology for eliminating foodborne pathogens and improving the safety of meats (Trindade, Mancini-Filho, & Villavicencio, 2010). It is approved for use in over 55 countries worldwide for various applications and purposes in a wide variety of foodstuffs (IAEA, 2009). The major advantages of irradiating meats are that it is a nonthermal process, it maintains the integrity of the products, and leaves no chemical residues. Also, the products can be treated after packaging, which prevents further cross-contami-

nation during postprocessing handling. However, the acceleration of oxidation and off-odour production caused by irradiation in meat products has been reported (Lim, Seol, Jeon, Jo, & Lee, 2008). The development of oxidative off-flavour has long been recognised as a serious problem during the holding or storage of meat products. Furthermore, oxidation leads to health disorders including cancerogenesis, therefore, enhanced oxidative stability is needed for maintaining the quality and safety of meat products (Arabshahi-D, Devi, & Urooj, 2007; Trindade et al., 2010).

The use of antioxidants is one of the major strategies for preventing lipid oxidation and may be effective in controlling and reducing the oxidation in irradiated meat products. Due to the fact that chemical additives may constitute a potential health hazard for consumers, interest in natural antioxidants and search on naturally occurring compounds that have antioxidant effect has been increased dramatically. Carrot is one of the important widely consumed root vegetable with high nutritional value due to its enriched healthy composition, such as phytonutrients and minerals and its juice is one of the most popular vegetable juices and often marketed as a health drink (Zhou, Wang, Hu, Wu, & Liao, 2009). Carrot, as well as its juice, is a good source of natural antioxidants especially carotenoids and phenolic compounds, having the highest carotenoid content among foods (Arabshahi-D et al., 2007; Hsieh & Ko, 2008; Soria, Sanz, & Villamiel, 2009). Carrot juice has a pleasant flavour and may exert an antioxidant activity in sausages without adverse effects on their acceptability. Therefore,

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the objective of this study was to evaluate the antioxidant activity of different concentrated carrot juices in gamma irradiated beef sausages and their acceptability during refrigerated and frozen storage.

#### 2. Materials and methods

#### 2.1. Carrot juice

Fresh carrots were obtained from a local vegetable market. Carrots were topped and tailed using a knife, washed with tap water and peeled using a hand peeler. The peeled carrots were rinsed under tap water and the juice was obtained using a juice extractor. The obtained juice bulk was divided into three appropriate portions. The first portion was kept unconcentrated, while the volumes of the second and third portions were reduced by 35% and 60%, respectively, at 35 °C with a rotary evaporator.

#### 2.2. Beef meat

Beef cuts (flank and forequarters) were separately excised from three beef carcasses at the butcher's shop (after 3 h of slaughtering and dressing) and used separately as replications for the preparation of samples (three separate trial replicates). After the removal of connective tissue, the obtained beef cuts were separately chopped into small pieces and ground using a meat grinder.

#### 2.3. Manufacture of sausages

Four main patches of beef sausages were prepared within each trial replicate. The first batch was formulated with water and ice according to the traditional formula, to serve as the control sausage samples, and consisted of the following ingredients: beef flank (65%), beef quarters (10%), dextrose (0.695%), sodium caseinates (0.794%), soya (0.993%), ecolin (0.894%), dried garlic (0.198%), tripolyphosphate (0.369%), sodium chloride (1.142%), water and ice (19.148%), nitrite and nitrate (0.01%), and spices (0.730%). Samples of the other three batches were formulated using the same ingredients with the replacement of water and ice and dextrose by 19.843% of unconcentrated carrot juice, carrot juice concentrated by 35% and carrot juice concentrated by 60%, respectively. Sausages were prepared as described by Heinz and Hautzinger (2007) and stuffed into a nature casings which were hand linked at about 15 cm intervals. Samples of the prepared sausages were aerobically packaged in polyethylene pouches which were sealed by heat, then samples of each of the prepared batch were subdivided into three appropriate groups and transported for irradiation treatments in an ice chest.

#### 2.4. Irradiation of samples

Packaged samples of each batch of the raw beef sausages were gamma irradiated at doses of 0.0, 3.0, and 4.5 kGy, the doses approved for treating raw meat products (USDA-FDA, 2003). Irradiation was carried out at room temperature using an experimental Co-60 source (providing a dose rate of 2.527 kGy/h) located at the National Center for Radiation Research and Technology, Nasr City, Cairo, Egypt.

#### 2.5. Storage and sampling

Irradiated and non-irradiated samples of each of the prepared beef sausages were divided into two parts that required for the refrigeration storage at  $4 \pm 1$  °C and frozen storage at -18 °C (except samples for the day zero analysis). The periodical sampling

for analysis was carried out at 3 and 15 days intervals for refrigerated and frozen samples, respectively. Frozen samples were thawed overnight at  $4 \pm 1$  °C before analysis.

## 2.6. Determination of total carotenoids and total phenolics in the carrot juice

The contents of total carotenoids in concentrated and unconcentrated carrot juice were determined spectrophotometrically as described by Zhou et al. (2009), while their contents of total phenolics were measured spectrophotometrically using the Folin–Ciocalteu colorimetric method according to Gao, Bjork, Trajkovski, and Uggle (2000).

#### 2.7. Measurements of oxidation

#### 2.7.1. Lipid oxidation

The extent of lipid oxidation in samples was assessed through the determination of peroxide value (PV) and thiobarbituric acid reactive substances (TBARS). For the determination of PV, lipids were extracted from samples under investigation using 2:1 chloroform/methanol solvent (Folch, Lee, & Sloane, 1957), then the PV was determined in the recovered lipids according to the Official Methods of AOCS (1998). Meanwhile, the contents of TBARS were determined in samples of beef sausages using the method of Alasnier, Meynier, Viau, and Gandmer (2000).

#### 2.7.2. Protein oxidation measurement

Protein oxidation as measured by the total protein carbonyl content was assessed using the 2,4-dinitrophenylhydrazine (DNPH) method as described by Reznick and Packer (1994). The carbonyl content was calculated by obtaining the spectra at 355–390 nm of the DNPH-treated samples. Protein concentration was determined by spectrophotometry at 280 nm using bovine serum albumin as standard and the amount of protein carbonyl content was expressed as nmol carbonyl/mg protein.

#### 2.8. pH and water activity

The pH was measured with a pH meter on a suspension resulting by blending 15 g of sausage sample with 150 ml of distilled water. The water activity  $(a_{\rm w})$  of samples was measured as described by Vos and Labuza (1974).

#### 2.9. Microbiological aspects

At time of withdrawal from refrigerated or frozen storage, 10 g aliquots of sausage samples were removed aseptically to prepare the initial 1/10 dilution which was serially diluted in 0.1% peptone water as needed. Then, colony forming units for total aerobic mesophilic and psychrophilic bacteria were determined by plating on plate count agar medium and incubation at 30 °C for 3 days and 7 °C for 7 days, respectively, while total molds and yeasts were enumerated on malt agar medium after incubation at 25 °C for 3-5 days (APHA, 1992). Enterobacteriaceae were counted on violet red bile glucose agar medium after incubation at 37°C for 20-24 h (Roberts, Hooper, & Greenwood, 1995). Enumeration of Listeria monocytogenes was carried out on Listeria selective medium after incubation at 35 °C for 24-48 h (Oxoid, 1998). Cultures were examined for typical colonies after 24 and 48 h incubation, while colonies presumptively identified as L. monocytogenes were confirmed by biochemical testing (Roberts et al., 1995). Staphylococcus aureus was counted using Baird-Parker RPF medium after incubation at 35 °C for 24-48 h (Oxoid, 1998) and confirmed by the coagulase test as described by Collins, Lyne, and Grange (1989). The detection of Salmonella was carried out using the most

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