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CHANGES ON LIPID PROFILE IN BEEF BURGERS PREPARED WITH ROSEMARY EXTRACT AND SUBMITTED TO E-BEAM PROCESSING

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ABSTRACT

Radiation processing has been employed in some countries as a mean of treatment to assure microbiological safety of meat and meat products. Use of antioxidants for preventing lipid oxidation has been applied in those products. The present study aimed at evaluating the protecting effects of rosemary extract on the lipid profile of irradiated beef burgers. The samples were prepared with 400ppm of rosemary extract, irradiated at doses 0, 3.5 and 7kGy, stored at –20ºC for 45 days and after this time, evaluated in regard to the oxidative stability of lipids (TBARS values) and lipid profile in a GC (Gas chromatography). Non-irradiated and non-rosemary extract samples were used as a control. TBARS values were of 0.3 and 1.1 mgTBARS/kg of sample for rosemary extract and control samples (without extract) irradiated at 3.5kGy, respectively. At 7kGy, TBARS values were of 0.6 and 1.3 mgTBARS/kg of samples for rosemary extract and control samples (without extract), respectively. Total saturated fatty acid (SFA), monounsaturated fatty acid (MUFA) and polyunsaturated fatty acid (PUFA) did not change in beef burgers, although they showed small differences between the batches, this differences were not significant ($P<0.05$). The amounts of *Trans* fatty acid increased significantly ($P<0.05$) only when used irradiation dose of 7kGy (0.86 g/100g of sample). These results showed that the rosemary extract can avoid the developing of lipid oxidation and the irradiation processing did not change lipid profile in beef burgers.

1. INTRODUCTION

The public expects that our food supply should be safer than it is today because food-borne illnesses continue to be a problem in a world public health. Over the years, changes in farming practices and food processing have increased the complexity of the food distribution chain, which has opened new areas where pathogens can be introduced. Consequently, the country's food safety system needs to be modernized. One option is to improve food safety through food irradiation. Radiation processing has been employed in some countries as a mean of treatment to assure microbiological safety of meat and meat products.

Irradiation of lipids induces the production of free radicals, which react with oxygen, leading to the formation of carbonyls, responsible for alteration in food nutritional and sensory characteristics (Chen et al., 2006). Free radicals can be defined as any chemical species having one or more unpaired electrons, and one of the major areas in which carbon free radicals and oxygen free radicals are involved, is in the oxidation of lipids. This is a process in foods and bulk lipids which leads to rancidity. Rancidity is the spoiled off-flavour obtained by subjective organoleptic appraisal of food (Hamilton et al., 1996). Beef burgers are made of ground beef and have high lipid content. Typical composition of ground beef is about 18% lipids and its fatty acids content is divided into 46% saturated, 51% mono-unsaturated and 3% poly-unsaturated (Johnson et al., 1994). The ability of unsaturated fatty acids, especially those with more than two double bonds and induced by irradiation processes, to rapidly oxidise, is important in regulating the shelf life of meat (rancidity and colour deterioration) (Wood et al., 2003).

In order to inhibit the development of oxidative reactions in meat products, natural and synthetic antioxidants have been commonly used in meat industry (Estéves and Cava, 2006). Antioxidants are one element of a collection of processes that retard free radical oxidation. Use of antioxidants for preventing lipid oxidation has been applied in those products. The term antioxidation includes all the processes that slow down or stop free radical oxidation (Thomas, 2000).

Recent studies have described the use of herbs or plant extracts as inhibitors of oxidative reactions. Naturally occurring antioxidants compounds have been preferably employed in meats because of their potential health benefits and safety compared with synthetic preservatives, such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT) (Ahn et al., 2007). The antioxidative effects of rosemary (*Rosmarinus officinalis*) extract (rosemary) in meat processing have been monitored in some researches and its effects are recognized (Nassu et al., 2003; Lee et al., 2005)

2. EXPERIMENTAL

2.1. Manufacture of beef burger

Ready-to-eat beef burgers were prepared in a pilot processing plant in a local food industry at Brazil with the addition of an antioxidant (Rosemary extract). Ground meats, antioxidants, and ingredients were mixed in a commercial mixer. Rosemary extract was added at a concentration of 400 mg/kg within the range suggested for hamburgers by the manufacturer. After mixing, the meat batter was placed into a commercial machine into templates of 10 cm diameter and 7 mm height. The burgers were then packed in air-permeable polypropylene bags, frozen to -20 ºC and this temperature was maintained during all time of experiment (45 days) include the irradiation processing. The whole procedure was standardized to keep the time and temperature between the processing steps equal for all burgers.

2.2. Treatment by irradiation process

The samples were irradiated in an electron-beam irradiator (Institute for Nuclear and Energy Research - IPEN). The radiation dose level was 7.0 and 3.5 kGy for frozen samples (-20 ºC ± 5). Dose rate was 7.86 kGy/s and 15.67 kGy/s for 3.5 and 7.0 kGy, respectively. Samples were stored post-irradiation at -20 °C ± 5°C for frozen beef up to 45 days.

2.3. TBARS measurement

Malonaldehyde (MDA) and other thiobarbituric acid reactive substances (TBARS) were determined using a method of Turner et al. (1954). The *K* value was calculated using TEP 1,1,3,3, - tetraethoxypropane (Sigma Chemical Co) as the standard and TBARS values were calculated by multiplying the absorbance values by the *K* value. Results are expressed as mg MDA/kg beef burgers.

2.4. Fatty acids profile by Gas Chromatography (GC)

Fatty acids methyl esters (FAMEs) were prepared by method of AOAC (2001) by alkaline hydrolyze followed by esterification. FAMEs was analysed using a gas chromatograph (GC 17A Shimadzu). Identification of FAMEs was based on retention times of reference compounds (Sigma). The quantification of fatty acids was carried out by using C13 as an internal standard. Results are expressed as g fatty acid 100 g^{-1} total fatty acid detected.

3. RESULTS

3.1. TBA values of beef burgers

The antioxidant effects of treatment with rosemary extract, as measured by TBARS, over 45 days of frozen storage (-20ºC) are shown in **Figure 1**. TBARS values for the control (No rosemary) rapidly increased even before radiation submitting. TBARS value for sample with rosemary extract was significantly lower than those for the control at three doses utilized. Compared to the control, the sample with rosemary extract had significantly lower TBARS values throughout storage. This result indicates that lipid oxidation was effectively retarded by rosemary extract during and immediately after irradiation.

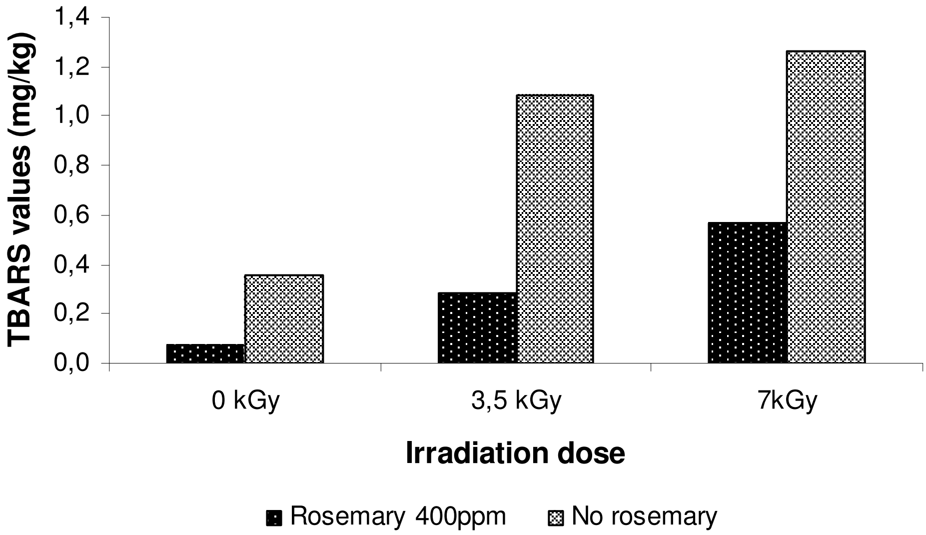

Figure 1. Lipid oxidation in beef burgers after 45 days conditioning at -20ºC.

3.2. Fatty acid composition

Fatty acids composition of beef burgers with rosemary extract and control samples after different doses of irradiation (0, 3.5 and 7 kGy) is presented in **Table 1 and 2**, respectively.

Typical composition of beef burgers is about 20% lipids and its fatty acid content is divided into about 50% saturated (SFA), 46% monounsaturated (MUFA), 3% polyunsaturated (PUFA) and 1% *trans* fatty acid.

Table 1. Fatty acid composition of beef burgers formulated with rosemary extract after 45 days of frozen storage.

Fatty acids	O kGy	3.5 kGy	7 kGy
Saturated	10,21±0,34 ^{A}	9,92±0,26 ^{A}	9,95±0,07 ^{A}
Monounsaturated	7,39±0,16 ^{A}	7,85±0,15 ^{A}	7,75±0,08 ^{A}
Polyunsaturated	0,67±0,16 ^{A}	0,58±0,10 ^{A}	0,50±0,02 ^{A}
Trans Fatty acids	0,80±0,04 ^{AC}	0,72±0,03 ^{A}	0,86±0,01 ^{BC}

$^{A\text{-}C}$ Means with different superscripts within a row are significantly different (p<0.05).

Table 2. Fatty acid composition of beef burgers control (No rosemary) after 45 days of frozen storage.

Fatty acids	O kGy	3.5 kGy	7 kGy
Saturated	10,06±0,03 ^{A}	9,90±0,01 ^{A}	10,07±0,08 ^{A}
Monounsaturated	7,66±0,02 ^{A}	7,82±0,02 ^{A}	7,64±0,04 ^{A}
Polyunsaturated	0,62±0,00 ^{A}	0,68±0,04 ^{A}	0,60±0,09 ^{A}
Trans Fatty acids	0,73±0,04 ^{A}	0,73±0,01 ^{A}	0,75±0,06 ^{A}

$^{A\text{-}C}$ Means with different superscripts within a row are significantly different (p<0.05).

4. DISCUSSION

2-Thiobarbituric acid (TBA) is a widely used compound owing to its reactivity, generally with carbonyl substances (aldehydes, ketones), although acids, esters, amides, sugars and pyrimidine compounds can also display reactivity with TBA (Guillén-Sans et al., 1998). Thus, although polyunsaturated fatty acid content did not decrease with irradiation process, TBARS values showed that has occur a high increase of oxidation in sample controls, without antioxidants, possibly due to oxidation of other food compounds, like, proteins, sugars, since that beef burger is complex moisture of nutrients. Although, TBA could be considered a general reagent for aldehydes, its main utilization has been in the recognition of the fat oxidation level, or the rancidity of fats. No rosemary control samples had significantly higher (P<0,05) TBARS values (0.4, 1.1 and 1.3 mgMDA/kg) than samples formulated with rosemary extract (0.1, 0.3 and 0.6 mgMDA/kg at the dose of 0, 3.5 and 7 kGy respectively) under frozen conditions of storage. This is in agreement with the study of Georgantelis et al (2007) that treated several samples, no irradiated, with different types of antioxidants, among them, rosemary extract at 200 mg/kg. They found that lipid oxidation in the controls was more intense compared to treated samples, reaching 4.0 mgMDA/kg after 180 days of storage. Since that TBARS values is considered as a indicators to rancidity in fat products, Verme and Sahoo (2000) indicate MDA concentrations between 1.0 and 2.0 mg/kg as a threshold values for rancidity, while Greene and Cumuze (1982) considered a TBARS range 0.6 – 2.0 mg/kg to be the minimum detectable level for oxidized flavour in ground beef. Accordingly, the rosemary samples in the present study would not be perceived by consumers up to 45 days of storage even over irradiation process at 7 kGy. Control samples did not show significantly difference between dose of 3.5 and 7.0 kGy, however, the samples with rosemary extract shows higher susceptibility to dose of irradiation, although both values keep under 1.0 mgMDA/kg.

The treatment of foods with radiation for conservation and microbial decontamination is an established method employed in several countries. In general, any modifications caused by treatment with doses up to 10 kGy were regarded to be harmless for human nutrition. On the other hand, chemical modifications in the lipid fraction of food by formation of radicals and hydrocarbon fragments are well known (Geibler et al., 2003). The effect of fatty acids on shelf life is explained by the propensity of unsaturated fatty acids to oxidize, leading to the development of rancidity as display times increase and the food passes through irradiation processes (Wood et al., 2003). Some of the fatty acids found in meat play important roles in metabolism. Polyunsaturated fatty acid such as linolei acid and arachidonic acids are of great nutritional importance being essential to the human diet as they cannot be synthesized within the body. The most susceptible site for free radical attack in a lipid molecule is at a double bond. The most affected lipids during irradiation are thus the polyunsaturated fatty acids that bear two or more double bonds available for reaction (Giroux and Lacroix, 1998). Natural fats and oils that are present in the human diet contain only small amounts of *trans* fatty acids. Animal sources of fats such as milk, dairy products, and beef usually contain between 2% and 4.5% of their total fat as *trans* fatty acids, which are generated during rumination and subsequently absorbed and stored in animal tissues (Yilmaz and Geçgel, 2006). In this present study, we observed that did not have changes in samples by radiation induction. Since that the samples were irradiated at 3.5 and 7 kGy under frozen conditions, the irradiation did not have energy enough to induce *trans* fatty acid formation. All samples showed about 0.8% *trans* fatty acids, although some variations can be seen among the values shown, any was significantly different. Yilmaz and Gecgel (2006) investigated the level of *trans* fatty acids in irradiated ground beef and found that irradiation increase trans fatty acids with increase of irradiation, but irradiation of samples was carried out under refrigerated conditions. Same results can be noted in the study of Brito et al (2002) under the same conditions of irradiation. In the present study, samples were irradiated under frozen condition. It can avoid the *trans* fatty acids formation because there was lower water activity and subsequently lower free radicals formation than that. The rate of fatty acids (SFA, MUFA and PUFA) did not change among different doses and different formulation over 45 days of storage, although they showed small differences between the batches, this differences were not significant ($P<0.05$).

5. CONCLUSIONS

These results showed that the rosemary extract can avoid the development of lipid oxidation and irradiation processing, under conditions of this experiment, did not change lipid profile in beef burgers. However, further studies are needed to determine the effective concentrations of natural antioxidants in meat products without adversely affecting the other characteristics of product.

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