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Consumer's evaluation of the effects of gamma irradiation and natural antioxidants on general acceptance of frozen beef burger

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ABSTRACT

The effect of addition of rosemary and oregano extracts on the sensory quality of irradiated beef burger was investigated. Batches of beef burgers were prepared with 400 ppm of rosemary or oregano extract and a group prepared with 200 ppm of synthetic butyl-hydroxytoluene (BHT)/butyl-hydroxy-anisol (BHA) was used as a control. Half of each formulation was irradiated at the maximum dose allowed for frozen meat (7 kGy). Samples were kept under frozen conditions (-20 °C) during the whole storage period, including during irradiation. Two analyses were performed after 20 and 90 days to verify the influence of the addition of the different types of antioxidants and the effect of irradiation and storage time on the acceptance of the product. Thirty-three and thirty-four untrained panelists were invited to participate in the first and second test, respectively. A structured hedonic scale ranging from 1 to 9 points was used in both analyses. BHT/BHA formulation obtained the highest score (6.73) and regarding the natural antioxidants, oregano received better acceptance (6.36). Irradiated samples formulated with oregano received a lower score, 6.03 in the first test and 5.06 in the second one, compared to the non-irradiated sample (6.36 and 5.79). In the second test (90 days), the sample formulated with BHT/BHA and which was irradiated received a higher score (6.59) when compared to the non-irradiated one (5.85). In both tests, the irradiated samples formulated with rosemary extract obtained a better score compared to the non-irradiated one, the scores being 5.00-3.82 and 5.00-3.76 in the first and second test, respectively. Our results allowed us to conclude that the natural antioxidants, rosemary and oregano extracts, present a good alternative for replacing synthetic additives in food industries, and that the irradiation process, in some cases, may help to enhance the sensory quality of food.

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1. Introduction

The efficacy of irradiation for eliminating pathogenic bacteria in food is well established. The Food and Drug Administration (FDA) has approved the use of ionizing radiation for refrigerated and frozen uncooked meat to control foodborne pathogens and to extend their shelf life (FDA, 1997). The meat industry has considered using this technology to improve the safety of their products and, if this was the case, how they could apply it (Olson, 1998).

Although there are conflicting data regarding the effect of irradiation on the sensory attributes and properties of meat (Thayer, 1990), efforts have been employed by researchers and food technologists to improve the sensory quality of irradiated

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products, as well as to create methods to reduce the loss of sensory attributes after irradiation. The loss of sensory quality is based on the fact that the development of off-odors and off-flavors in irradiated meat can be affected by a number of factors, including temperature, environment within the package, packaging material, radiation dose, post-irradiation storage time, types of ingredients added to the products, fat content (since lipids are more susceptible to oxidation caused by ionizing radiation), and the conditions under which the meat was irradiated. In order to minimize the development of objectionable off-odor and off-flavors, it has been recommended to irradiate meat and its sub-products in the frozen state at the approved doses of 3.5 and 7 kGy for refrigerated and frozen meat, respectively (FDA, 1997).

The major cause of loss of sensory quality in meat and its products is lipid oxidation. Lipid oxidation is a process that occurs by the action of free radicals, induced by irradiation, on molecules of unsaturated fatty acid, not only polyunsaturated fatty acids like

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linoleic (C18:2), linolenic (C18:3), and arachidonic (C20:4) acids, but also monounsaturated fatty acids like palmitoleic (C16:1) and oleic (C18:1) acids. Polyunsaturated fatty acids are more susceptible to radiation-mediated oxidation than saturated fatty acids since the energy needed to cause homolitic breakdown of C-H double bonds (~60 kcal/mol) is lower than the energy required to cause the same effect on C-H single bonds present on saturated chains (~100 kcal/mol). Lipid oxidation occurs not only as consequence of the irradiation process, but also under many other situations, including auto-oxidation, which affects oils, fats, and oil-based foods (Fennema, 2007).

In order to avoid, inhibit, or retard lipid oxidation in meat, meat products, and other oil-based products, and to extend the shelf life of these products, food industries have been using antioxidants. Antioxidants are substances that counteract the free radicals, and by doing so they prevent and reduce the adverse damage by inactivating the free radicals before these can react with biologic targets, thus avoiding chain reactions and the activation of oxygen to highly reactive products. In the irradiation process, the antioxidants can also function as scavengers of free radicals derived from the hydrolysis of water by ionizing radiation and, as a result, prevent further formation of free radicals and protect lipid molecules from ionizing rays. The largest class of antioxidants is that containing polyphenols. Polyphenols consist of phenolic acids and flavonoids (Venkat Ratnam et al., 2006). According to their sources, the antioxidants can be classified in two major categories: synthetic and natural. Butyl-hydroxy-anisol (BHA), butyl-hydroxytoluene (BHT), and terc-butyl-hydroquinone (TBHQ) are synthetic additives, which contain a phenolic structure similar to that contained in the polyphenols derived from natural sources, such as herbs and spices, and which are classified as natural antioxidants. Although many other natural antioxidants have been investigated, rosemary and oregano are probably the most widely investigated natural antioxidant systems and these two are not only used in meat products, but also in general foods. Previous studies demonstrated that antioxidants could reduce the effects of oxidation on irradiated meat (Nam and Ahn, 2003) and that, in some cases, they may trigger a differential taste. On the other hand, studies have also demonstrated possible undesirable effects on health caused by synthetic additives. For this reason, consumers have given preference to natural additives in food (Hocman, 1988; Ito et al., 1986; Williams and Iatropoulos, 1996; Williams et al., 1999).

The addition of herbs and spices to meat products depends, of course, on their sensory compatibilities with meats as was reported almost 20 years ago (Rhee, 1987). This has resulted in several researches towards using extracts of spices and herbs, and some alternatives to reduce the effects of the addition of these natural substances have also been proposed that are beneficial in terms of chemical protection and the consumer's sensory perception. The major characteristics of phenolic compounds in plants and agroindustrial by-products have been reviewed in a recent paper by Balasundram et al. (2006).

Therefore, the objective of this study is to evaluate the general consumer acceptance of rosemary and oregano extracts as natural antioxidants added to beef burgers that are submitted to high and maximum ionizing radiation dose allowed for meat in frozen conditions (7 kGy).

2. Materials and methods

2.1. Materials

The leaves of oregano (*Origanum vulgare* L.) were purchased from a local market. Rosemary extracts (GuardianTM) and BHT/

BHA blend (GrindoxTM) were obtained from Danisco S/A (São Paulo, BR); β -carotene, linoleic acid puriss. p.a., and Tween 40 were obtained from Sigma-Aldrich Chemical Corporation (St. Louis, MO, USA). Ethyl alcohol 95%, methyl alcohol, ethylic ether p.a., chloroform p.a., and all reagents were of analytical grade.

2.2. Extract preparation

Oregano extracts were obtained from sequential extraction using solvents with different polarities, beginning with ethylic ether, followed by ethylic alcohol and distillated water as reported by Mancini-Filho et al. (1998). For formulation with rosemary, commercial extracts were used since they are available to the food industry.

2.3. Antioxidant activity

In order to better define the dose that would be added to the samples, the antioxidant capacity was determined by the β -carotene bleaching method so as to estimate *in vitro* the antioxidative potential of the herbs used in this study. The test was carried out following the spectrophotometric method developed by Marco (1968) and modified by Miller (1971), which is based on the ability to decrease the oxidative bleaching on β -carotene in a β -carotene/linoleic acid emulsion. This is a co-oxidation system of substrates in which a spectrophotometric assay based on discolouration (oxidation) of β -carotene induced by oxidative degradation products of an unsaturated fatty acid (linoleic acid) is carried out. Different quantities of extracts (see prepared solutions in Table 3), based on gravimetric measures and expressed in parts per million (ppm), were added to 5 mL of β -carotene solution containing linoleic acid: 20 μ L β -carotene solution (20 mg/mL)+1 mL of chloroform, 40 mg of linoleic acid. and 530 mg of Tween 40 as an emulsifier, and were removed by evaporation with nitrogen. Furthermore, about 120-150 mL of distillated water treated with O₂ for 30 min was added. The final solution was diluted up to an optical density between 0.6 and 0.7 at 470 nm. This system was kept at 50 °C and spectrophotometric measures of absorbance were done after 15, 30, 45, 90, 105, and 120 min of incubation in a water bath at 50°C, at 470 nm. Each sample was read against an emulsion prepared as described, but without β -carotene (blank). The absorbance of the mixtures at each time point was read with a spectrophotometer, and the absorbance measured was subtracted from that of the corresponding sample. The degradation rate (D) of β -carotene was calculated by first-order kinetics:

$D_{\text{carotene}} = [\ln(A_0/A_t)]/t$

where A_0 is the absorbance of the sample–the absorbance of the blank sample at time 0 (absorbance was read immediately after the addition of alkaloid solutions). A_t is the absorbance of the sample–absorbance of the blank sample at time t, and t = 15, 30, 45, 90, 105, or 120 min of incubation at 50 °C.

In all, 100 or 200 ppm of extracts were added to 5 mL of β carotene emulsion and treated as the corresponding sample; A_0 is the absorbance of the control sample at time 0, and A_t the absorbance of the control sample at time *t*. Antioxidant activity (*A*) was expressed as a percentage of inhibition relative to the control using the equation

$$A = \frac{d_{\text{control sample}} - d_{\text{sample}}}{d_{\text{control sample}}} \times 100$$

2.4. Sample manufacturing

Ready-to-cook beef burgers were prepared with the addition of an antioxidant (Table 1) based on industrial data that are currently used at a local processing industry in the city of São Paulo. Ground meats and ingredients were divided into three batches. Oregano and rosemary extracts alone (400 ppm each) and BHT/BHA alone (200 ppm) were added to the three batches. Ground meats, antioxidant, and ingredients were mixed in a commercial mixer and molded in an industrial molder (belonging to a local industry). The beef burgers were then aerobically packaged in polyethylene bags and were held under frozen conditions (-18 to -20 °C) until the irradiation process.

2.5. Gamma irradiation and storage

Gamma irradiation was carried out in a ⁶⁰Co semi-industrial irradiator, installed at the Institute for Energy and Nuclear Research (*Instituto de Pesquisas Energéticas e Nucleares*—IPEN, São Paulo, Brazil). The applied radiation dose levels were 0 (control) and 7 kGy (\pm 10%). Half of each formulation was irradiated. The equipment operated at a dose rate of 3 kGy/h (\pm 10%). During the whole irradiation period, the samples were held frozen (-18 to -20 °C) with dry ice inside thermal boxes. Harwell Amber 3042 dosimeters were used for the measurement of the radiation dose (\pm 10%). To minimize variations in the radiation dose absorption, the thermal boxes that contained the samples were turned 180° halfway through the procedure. After irradiation, the beef burgers were stored in a freezer at -20 °C during the experimental period (90 days).

2.6. Sensory evaluation

Thirty-three and thirty-four students of the Department of Food Science, Faculty of Pharmaceutical Sciences, University of São Paulo were selected as panelists on the level of consumers and they conducted sensory evaluations twice, that is, on day 20 and day 90 of storage, respectively. Samples of approximately 50g were cooked at an industrial kitchen in the Laboratory of Sensory Evaluation of the Faculty of Food Engineering at the University of São Paulo and presented to the panelists. The samples were prepared as follows: the temperature of the grill was kept at 180 °C and the beef burger samples were fried for 2 min plus one extra minute for each side to turn golden brown, after which they were kept inside a thermal box. Samples were served to the panelists as follows: each beef burger was cut into four pieces and accompanied by a glass of water. Water and salt cracker was available to the panelists to rinse their palates between one sample and the other.

An affective panel was used for measuring consumer reactions to a particular product (beef burgers) in terms of acceptance/ preference. A total of six samples were presented to the panelists, three irradiated and three non-irradiated. The panelists were asked to give scores to the samples using a structured hedonic scale ranging from 1 to 9, 1 being extremely disliking and 9 being extremely liking. In both analyses (day 20 and day 90), a total of 67 consumer panelists volunteered to participate in response to an invitation sent to students and workers of the University of São Paulo, such that there were 33 and 34 panelists in the first and second test, respectively. Panelists were not compensated and they were previously informed that the study would include samples that had been irradiated. Beef burgers used for consumer evaluation came from all six batches. In two sessions separated by 70 days (day 20 and day 90), consumers evaluated the samples in terms of irradiation dose and the type of antioxidant (each sample was one fourth of a hamburger). They were given the samples (one fourth of a patty) one at a time (identified by random threedigit codes). It is important to emphasize that the first evaluation happened after 20 days of production to simulate the commercial interval that occurs from production to when the products are sold to final consumers. Sensory evaluation of this product was approved by the ethics committee of the Faculty of Pharmaceutical Sciences of the University of São Paulo and all participants were notified about the irradiation process and additives used on the samples.

2.7. Statistical analysis

Means and standard deviations (SDs) from the measurements within a batch were obtained from all analytical experiments. Results from the experiments, including the score given by panelists, were used as variables and analyzed by using a one-way analysis of variance (ANOVA) using GraphPad Prism 4.03 for Windows (GraphPad Software, San Diego, CA, USA) in order to assess the effect of the irradiation and addition of the antioxidants on sensory quality of beef burgers and preference differences among the samples. When statistically significant differences were found, Tukey tests were performed. Statistical significance was set at p < 0.05. A box plot of the scores given by panelist is shown for verification of the characteristics of their distribution in a hedonic scale.

3. Results and discussion

3.1. Antioxidant activity in β -carotene/linoleic acid system

The capacity to inhibit oxidation in the β -carotene/linoleic acid system submitted to oxidant conditions of both the commercial extract of rosemary Danisco[®] and aqueous extract of oregano, when used alone and in combination with each other, and also the combination of natural extracts and synthetic BHT/BHA, which was used as parameter of comparison, are presented in Table 2. It was observed that rosemary extract has a high antioxidant potential at a concentration of 100 ppm (77.67%) and also at a concentration of 200 ppm (85.62%). A better performance presented by rosemary extract was observed when it was used in combination with BHT/BHA at a concentration of 100 ppm each

Table 1

Addition of antioxidants for the preparation of experimental treatments of beef burgers.

Formulations	Beef (%)	Bovine fat (%)	Iced water (%)	Salt (%)	BHT/BHA (ppm ^a)	Rosemary ^b (ppm)	Oregano ^c (ppm)
BHT/BHA	70.0	20.0	8.0	2.0	200	-	-
Rosemary extract	70.0	20.0	8.0	2.0	-	400	-
Oregano extract	70.0	20.0	8.0	2.0	-	-	400

^a ppm: *parts per million* on the basis of total mass (mg kg $^{-1}$).

^b Commercial extract.

^c Aqueous extract.

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Antioxidant activity (%) in β-carotene/linoleic acid system.

Solutions	Test 1—rosemary (%) ^a	Test 2—oregano (%) ^a
Control	_	_
BHT/BHA 100 ppm ^b	94.48	97.38
BHT/BHA 200 ppm	98.13	95.58
Rosemary or oregano 100 ppm	77.67	60.63
Rosemary or oregano 200 ppm	85.62	75.15
Rosemary 50 ppm+BHT/BHA 50 ppm	86.50	-
Rosemary 100 ppm+BHT/BHA 100 ppm	89.71	-
Rosemary 50 ppm+oregano 50 ppm	74.66	61.49
Rosemary 100 ppm+oregano 100 ppm	80.42	72.83
Oregano 50 ppm+BHT/BHA 50 ppm	-	95.06
Oregano 100 ppm+BHT/BHA 100 ppm	-	97.38

^a All analyses were done in triplicate.

^b ppm: parts per million on the basis of total liquid (mg L^{-1}).

(89.71%). It was observed that aqueous extract of oregano presented a lower antioxidant potential compared to the commercial extract of rosemary, that is, the percentage of inhibition was 75.15% at a concentration of 200 ppm. However, the antioxidant ability of oregano in combination with BHT/BHA at a concentration of 100 ppm each was 97.38%, demonstrating that the occurrence of a synergistic effect between these products is possible.

Fig. 1 shows the kinetics of protection against oxidation (antioxidant capacity) of both commercial extract of rosemary (A) and aqueous extract of oregano (B), used alone and in combination with each other, and also with BHT/BHA in β -carotene/linoleic acid system. The β -carotene/linoleic acid system is one of the most used assays to evaluate antioxidant capacity of a variety of products. This system presents simplicity and sensibility as a method, although it is not very specific (other oxidant substances may interfere in the test). According to Silva et al. (1999), as the co-oxidation of β -carotene is normally done in

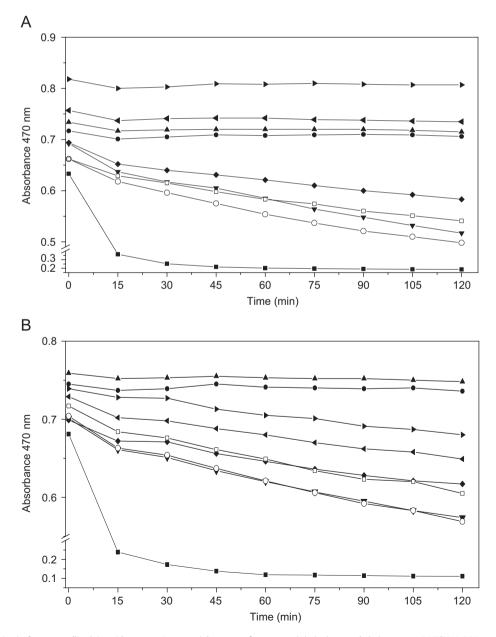


Fig. 1. Antioxidant activity in β-carotene/linoleic acid system. Commercial extract of rosemary (A): (■) control; (►) oregano+BHT/BHA 200 ppm; (◀) oregano+BHT/BHA 100 ppm; (▲) BHT/BHA 200 ppm; (●) BHT/BHA 100 ppm; (♦) oregano 200 ppm; (♥) oregano 100 ppm; (□) oregano+rosemary 200 ppm; (○) oregano+rosemary 100 ppm. Aqueous extract of oregano (B): (■) control; (►) rosemary+BHT/BHA 200 ppm; (◀) rosemary+BHT/BHA 100 ppm; (▲) BHT/BHA 200 ppm; (●) BHT/BHA 100 ppm; (♠) rosemary 200 ppm; (♥) rosemary 100 ppm; (●) rosemary+oregano 200 ppm; (○) rosemary+oregano 100 ppm.

an emulsified medium, it is common to produce low reproducibility. For this reason, this assay may be useful as a screening test for selection and guidance concerning antioxidative capacity *in vitro*, like the way it has been used in the present study.

Isolated compounds of rosemary have shown antioxidant effects when they were evaluated in emulsion, vegetable oil system, pork fat, and many other food matrices. The commercial extract of rosemary presented higher antioxidant ability than aqueous extract of oregano in our experiments. Similar results were found by Cintra (1999), who compared the antioxidant activity of rosemary extract and various others spices, among them oregano extract. Although the values found by Cintra (1999) were lower than those obtained in our study, the general performance was similar since a variation of the values is noted due to the place of harvest, the time, and storage conditions, and mostly due to the type of extraction to which the samples were submitted.

Previous studies have been carried out with a commercial version of extract of rosemary on food, but they did not report values related to antioxidant activity in oxidation system (Esteves and Cava, 2006; Ahn et al., 2007; Lee et al., 2005). Recent works on rosemary indicate alternative ways of extraction that attempt to reduce the loss of bioactive compounds present in this spice. Even under these conditions of extraction, when these extracts were tested in β -carotene/linoleic acid system to evaluate their antioxidant activity, they presented similar results (Carvalho-Junior et al., 2005). In our experiment, rosemary and oregano extracts were submitted to the same assay for comparison purposes. The combination (synergism) results between rosemary and BHT/BHA are in accordance with the results of Basaga et al. (1997), who evaluated the synergistic effect of rosemary and BHT on oxidative stability of soy oil.

The aqueous extract of oregano presented a lower antioxidant capacity than that of rosemary, and both rosemary and oregano alone presented a lower antioxidant capacity than that of synthetic BHT/BHA even though rosemary and oregano can also be considered to have a high potential since they are derived from a natural source. Almeida-Dória and Regitano-D'arce (2000) found similar results when they tested rosemary and oregano in β -carotene/linoleic acid system and compared them to BHT/BHA, and also between oregano and rosemary, where the latter presented a better performance even though it was used at a higher concentration, 500 and 1000 ppm for rosemary and oregano, respectively. Maybe the authors decided to use higher concentrations because of the usage of alcohol extract, which, according to Cintra (1999), has reduced quantity of phenolic compounds.

Nowadays, several other studies have emphasized the extraction technique of bioactive compounds from spices and herbs to maximize their use as antioxidants. Among the different extraction techniques, subcritical water extraction (Rodriguez-Meizoso et al., 2006) and supercritical fluid extraction (Carvalho-Junior et al., 2005; Cavero et al., 2006) can be cited. Other previous works have reported and proved the antioxidant potential of spices, including their use on different food matrices, such as pork sausage (Sebranek et al., 2005), beef patties (Formanek et al., 2001), beef meat (Georgantelis et al., 2007), cooked beef meat (Ahn et al., 2007), pork hamburgers (Lee et al., 2005), and liver pâté (Estéves et al., 2007).

In our experiment, the aim of performing this assay was to establish parameters referent for addition of extracts on the samples. Based on these results, it was decided that 200 ppm of BHT/BHA, 400 ppm of rosemary, and the same amount of oregano extract would be used since the threshold for using natural additives in food is that which does not interfere with the sensory and nutritional quality of food.

3.2. Sensory evaluation

Sensory evaluation is defined as the science of judging and evaluating the quality of a food by the use of the senses, i.e. taste, smell, sight, touch, and hearing. Sensory testing has been developed into a precise, formal, and structured methodology that is continually being updated to refine existing techniques. Sensory evaluation is used as a practical application in product development by aiding in product matching, improvements, and grading. Simply stated, sensory evaluation is divided into two methods, subjective and objective testing. Subjective tests involve objective panelists, while objective testing employs the use of laboratory instruments with no involvement of the senses (Meilgaard et al., 1999). One such subjective test is the affective panel with use of the hedonic scale method. This rating scale method measures the level of the liking of foods, or any other product where an affective tone is necessary. This test relies on people's ability to communicate their feelings of like or dislike. Hedonic testing is popular because it may be used with untrained people as well as with experienced panel members, where the best results are obtained with an untrained panel (Amerine et al., 1965). A minimum amount of verbal ability is necessary for reliable results (OMahony, 1986). In hedonic testing, samples are presented in succession and the subject is asked to decide how much he likes or dislikes the product and to mark the scales accordingly. The nature of this test is its relative simplicity. The instructions to the panelist are restricted to procedures, and no attempt is made at a direct response. The subject is allowed, however, to make his own inferences about the meaning of the scale categories and determine for himself how he will apply them to the samples. The scales are provided grouped together on a single page, where one scale is present for each sample being tested. The hedonic scale is anchored verbally with nine different categories ranging from extremely liking to extremely disliking. These phrases are placed on a line-graphic scale either horizontally or vertically. Hedonic ratings are converted to scores and treated by rank analysis or ANOVA (ASTM, 1968).

General acceptance of beef burgers by sensory evaluation reported as scores from 1 to 9 is shown in Table 3. Similar profiles of acceptance by the panelists were observed twice, that is, on both storage days (20th and 90th days). Campo et al. (2006) reported that it is very difficult to establish the exact moment at which a food must be rejected due to lipid oxidation based only on sensory perceptions, because this perception depends on personal feelings, which can vary for many reasons, mainly personal experience with the analyzed product. In this study, the samples were evaluated before lipid oxidation reached higher values; thus the different types of antioxidants and effects from irradiation on beef burgers could be noted by the panelists.

It was observed that within 20 days of storage, that is, soon after their production, the non-irradiated samples formulated with BHT/BHA received a higher score (6.73) than the

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Scores attributed to general acceptance in the sensory evaluation.

Samples	20 days	90 days
BHT/BHA 200 ppm—0 kGy	6.73^{a}	5.85^{ac}
BHT/BHA 200 ppm—8 kGy	6.55^{a}	6.59^{a}
Oregano 400 ppm—0 kGy	6.36^{a}	5.79^{ad}
Oregano 400 ppm—8 kGy	6.03^{ab}	5.06^{cd}
Rosemary 400 ppm—0 kGy	3.82^{c}	3.76^{b}
Rosemary 400 ppm—8 kGy	5.00^{bc}	5.00^{cd}

Note: different superscript letters in row within time storage mean significant difference ($p \le 0.05$).

irradiated one (6.55). On the contrary, after 90 days of storage, the irradiated sample obtained a higher score (6.59) than the non-irradiated one (5.85), but both did not show significant differences (p > 0.05).

Although the aqueous extract of oregano showed lower antioxidant ability than the other types of antioxidants when it was evaluated, directly in the food (data not shown), this formulation obtained a better sensory acceptance compared to the sample formulated with rosemary extract, and this difference was significant ($p \leq 0.05$) in both storage periods. This lower acceptance of the sample formulated with rosemary extract is partly due to the strong residual taste of this spice. This hypothesis is confirmed when it is observed that in both testing periods, the rosemary sample obtained a lower score compared to the oregano sample, being that the difference in the first test was not significant (p > 0.05) and the difference in the second one was significant ($p \le 0.05$).

In Fig. 2, box plot graphs of the score distribution that were given to the samples by the panelists are shown. A box plot is a univariate presentation of the data distribution. The length of the box represents the range for the middle 50% of the data. A horizontal line in the center is drawn at the median (50th percentile). A vertical line is drawn from the box to the farthest data point. Potential outliers are displayed as separate dots if the distance between these data points and the box is larger than a selected value (1.5 quartile). It is important to emphasize that in our test, the panelists were not provided with a reference sample to anchor their judgments; so the scores were given based on the preference of each participant, and from the graph it is possible to see that there was a general concordance among the

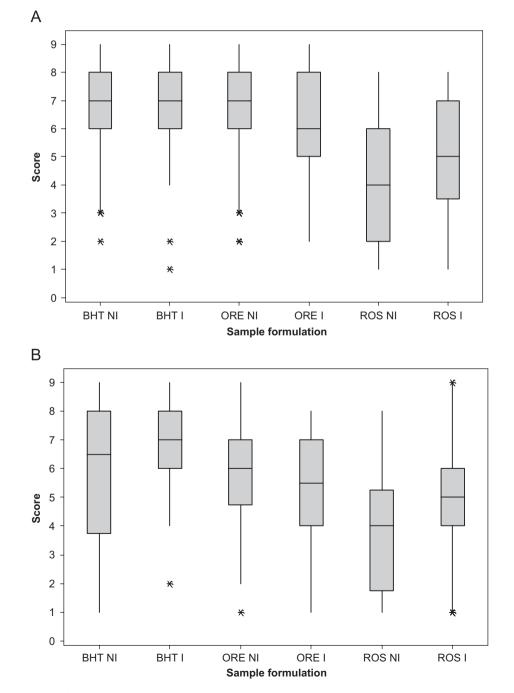


Fig. 2. Box plot of score distribution: 20 days (A) and 90 days (B). ROS, rosemary; ORE, oregano; NI, non-irradiated; I, irradiated.

participants; that is, at least 50% anchored their judgments in the strict group scores, for example, 50% of the scores given to the non-irradiated sample formulated with oregano extract (ORE NI—Fig. 2A) were centered between 6 and 8 on the hedonic scale. In the first test period, samples formulated with BHT/BHA and oregano, irradiated or not, obtained a good score distribution with 50% above 5. However, samples formulated with rosemary obtained lower scores. Interestingly, in these samples, the irradiated ones obtained a good acceptance. It is possible that the panelists did not associate the taste of rosemary with beef meat, since in Brazil this herb is more commonly used in fishbased foods, and we believe that when these samples were irradiated they lost the rosemary characteristic taste and became more acceptable to consumers. For this reason, the production of deodorized rosemary extracts could be an option to solve this undesirable influence in the flavor quality.

Although the scores given by the panelists to all the tested samples were low, this can be explained by the fact that they were non-trained panelists; thus the consumer's attitude towards the product was simulated, but considering that hamburgers are usually ingested with mayonnaise, tomato sauce, and/or mustard sauce, the taste could have been underestimated (Chirinos et al., 2002). In our experiment, condiments or other types of additives that could overcome the oxidative taste were not used. Kim et al. (2002) also performed a sensory evaluation to verify the aroma of irradiated and non-irradiated beef meat at a dose of 3 kGy using a hedonic scale ranging from 1 to 7 and verified that the score given by the panelists was also very low, about 2.1 and 2.6 for irradiated and non-irradiated beef, respectively, and without any significant differences (p>0.05). However, it could be observed that consumers have difficulty evaluating these types of products that are normally consumed with other condiments and ingredients. Our results are in accordance with previous studies performed with meat-derived products and both natural and synthetic antioxidants. Lee et al. (2005) compared the addition of rosemary extract with BHA and they also used a control without antioxidants. They observed that the samples had fewer differences in global acceptance, but this study was carried out soon after the irradiation process and did not evaluate the storage time, considering that storage time has a great influence on the sensory quality due to the secondary effects of free radicals and other derived substances. Byun et al. (2000) irradiated beef sausage and evaluated the sensory quality of these products and observed that no alterations occurred between irradiated and non-irradiated beef sausages. The reports in the scientific literature related to irradiation effects on sensory characteristics of foods are controversial. Kregel et al. (1986) did not observe significant difference (p>0.05) in sensory attributes between irradiated and non-irradiated burgers. Murano et al. (1998) also did not find any significant alterations between non-irradiated frozen burgers and those irradiated at 2 kGy. On the contrary, Montgomery et al. (2003) used a dose of 2 kGy and observed an offflavor formation in all irradiated burgers compared to the nonirradiated ones. More recently, Moura (2004) also observed a significant difference ($p \leq 0.05$) between irradiated samples of hamburgers using a dose of 3 and 7 kGy when compared to the control samples. In the present work, significant differences between irradiated and non-irradiated samples were not found (p>0.05), except for the samples formulated with rosemary extract. Therefore, when this sample was irradiated it presented a higher acceptance compared to the non-irradiated one. According to Esteves and Cava (2006), although the use of spices and essential oils on meats and fat products is a common practice and a tendency in the industrial routine, their effects may be undesirable depending on the concentration of substances and also on the chemical-organoleptic characteristics of products. Thus, the decision to use a specific quantity of natural extracts for the purpose of antioxidant protection must be carefully studied, taking into consideration that each food product has a particular sensory characteristic.

In some specific cases, the irradiation process may help to enhance the sensory perceptions of food, mainly when natural extracts containing volatives that cause off-flavor are added. It is observed in Table 3 that the samples formulated with rosemary obtained a higher score (5.00–3.82) when irradiated compared to the non-irradiated one (5.00–3.76) in both tests. It is possible that the irradiation process helped to rapidly volatilize the substances that trigger a non-characteristics aroma in the food.

Currently, deodorized extracts of rosemary that are very useful for products that do not have affinity to the residual taste of this spice are available to food industries that search for other natural alternatives for their products. Moreover, alternative ways of extracting antioxidant compounds from spices and herbs, aiming to enhance their bioactivity and eliminate possible residual taste, should be explored by food technologists.

4. Conclusion

Beef burgers formulated with BHT/BHA obtained higher scores in the sensory evaluation when compared to other formulations with natural antioxidants, but between rosemary and oregano extract, the latter obtained a better general acceptance by the panelists under the conditions of this experiment.

Consumer's preference was not altered by ionizing radiation since the score given to the samples by the panelists did not show any significant differences (p > 0.05), except for the sample formulated with rosemary, where irradiation seems to rise the score in the two analyses performed. Moreover, the order of preference also was similar in the both tests.

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