Oxidative stability of goat hamburger added with black sesame extract

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Abstract
Goat meat has taken a prominent role for the Northeast region of Brazil, and meat products such as hamburgers are a way of adding value to this raw material. Thus, the objective of this work was to prepare goat hamburgers and evaluate the effect of black sesame extract as a natural antioxidant during 40 days of storage. Four formulations were made: F1 - with bacon and no added antioxidant; F2 - with bacon and addition of 0.5% of black sesame extract; F3 - with soybean oil and without addition of antioxidant and F4 - with soybean oil and addition of 0.5% of black sesame extract. The hamburgers were subjected to centesimal composition analysis and evaluation of oxidative stability for 40 days. The treatments did not differ in terms of moisture and ash contents, with average values of 69.55 and 1.34%, respectively. The lipid content is in accordance with Brazilian legislation (maximum 23%), where it was possible to observe that the addition of black sesame extract, as it is an oilseed, provided an increase in the lipid content between F1 (10.96%) and F2 (11.97%) and between F3 (12.89%) and F4 (15.00%). Regarding the oxidative stability study, the use of saturated fat (bacon) and the action of black sesame as a natural antioxidant provided a lower oxidation of hamburgers, observing values of 11.44; 3.64; 18.07 and 16.04 mg MDA/kg of sample for F1, F2, F3 and F4, respectively. Thus, the black sesame extract was efficient for oxidative control of goat hamburgers.

Keywords: Caprinoculture; Meat product; Natural antioxidant; Oxidation in meat.

Resumo
A carne caprina tem assumido papel de destaque para a região Nordeste do Brasil e os derivados de carne, como o hambúrguer, são uma forma de agregar valor a essa matéria-prima. Assim, o objetivo deste trabalho foi preparar hambúrguer de cabra e avaliar o efeito do extrato de gergelim preto como antioxidante natural durante 40 dias de armazenamento. Foram feitas quatro formulações: F1 - com bacon e sem antioxidante adicionado; F2 - com bacon e adição de 0,5% de extrato de gergelim preto; F3 - com óleo de soja e sem adição de
antioxidante e F4 - com óleo de soja e adição de 0,5% de extrato de gergelim preto. Os hambúrgueres foram submetidos à análise da composição centesimal e avaliação da estabilidade oxidativa por 40 dias. Os tratamentos não diferiram quanto aos teores de umidade e cinzas, com valores médios de 69,55 e 1,34%, respectivamente. O teor de lipídios está de acordo com a legislação brasileira (máximo 23%), onde foi possível observar que a adição do extrato de gergelim preto, por se tratar de uma semente oleaginosa, proporcionou aumento no teor de lipídios entre F1 (10,96%) e F2 (11,97%) e entre F3 (12,89%) e F4 (15,00%). Em relação ao estudo da estabilidade oxidativa, o uso da gordura saturada (bacon) e a ação do gergelim preto como antioxidante natural proporcionou uma menor oxidação dos hambúrgueres, observando-se valores de 11,44; 3,64; 18,07 e 16,04 mg MDA/kg de amostra para F1, F2, F3 e F4, respectivamente. Dessa forma, o extrato de gergelim preto foi eficiente no controle oxidativo de hambúrgueres caprinos.

Palavras-chave: Caprinocultura; Produto cárneo; Antioxidante natural; Oxidação na carne.

Resumen
La carne de cabra ha tomado un papel destacado para la región Nordeste de Brasil, y los productos cárnicos como las hamburguesas son una forma de agregar valor a esta materia prima. Así, el objetivo de este trabajo fue preparar hamburguesas de cabra y evaluar el efecto del extracto de sésamo negro como antioxidante natural durante 40 días de almacenamiento. Se hicieron cuatro formulaciones: F1 - con tocino y sin antioxidantes añadidos; F2 - con tocino y adición de 0,5% de extracto de sésamo negro; F3 - con aceite de soja y sin adición de antioxidante y F4 - con aceite de soja y adición de 0,5% de extracto de sésamo negro. Las hamburguesas se sometieron a análisis de composición centesimal y evaluación de estabilidad oxidativa durante 40 días. Los tratamientos no difirieron en términos de contenido de humedad y cenizas, con valores promedio de 69,55 y 1,34%, respectivamente. El contenido de lípidos está de acuerdo con la legislación brasileña (máximo 23%), donde se pudo observar que la adición de extracto de sésamo negro, por ser una oleaginosa, proporcionó un aumento en el contenido de lípidos entre F1 (10,96%) y F2. (11,97%) y entre F3 (12,89%) y F4 (15,00%). En cuanto al estudio de estabilidad oxidativa, el uso de grasas saturadas (tocino) y la acción del sésamo negro como antioxidante natural proporcionaron una menor oxidación de las hamburguesas, observándose valores de 11,44; 3,64; 18,07 y 16,04 mg MDA / kg de muestra para F1, F2, F3 y F4, respectivamente. Así, el extracto de sésamo negro resultó eficaz para el control oxidativo de las hamburguesas de cabra.
Palabras clave: Caprinocultura; Producto cárnico; Antioxidante natural; Oxidación en la carne.

1. Introduction

With the growing search for healthy foods and the appreciation of regional culture, goat meat has been showing great progress in the development of new meat products, due to its characteristics of high protein value and low levels of fat, cholesterol and calories, compared to beef and pork, attracting the market of demanding consumers, and the preparation of meat products, such as hamburgers, is a great alternative to add value and facilitate their entry into the consumer market (Madruga 2005; Madruga et al. 2007).

Hamburger is an industrialized meat product obtained from the ground meat of butchery animals, whether or not added with adipose tissue and ingredients, molded and subjected to an appropriate technological process, being called Hamburger followed by the name of the animal species (Brasil, 2000).

The addition of fats, both solid and liquid, is normally used to guarantee desirable sensory characteristics, such as softness, flavor and palatability; however, these same lipid sources make the product much more susceptible to oxidation processes, which in turn can lead to reduced product shelf life (Oliveira, 2012). Thus, a very common practice to increase the stability of lipids in foods is the addition of antioxidants. Due to the lower cost and efficient action, the antioxidant compounds most used in the food industry are polyphenols of synthetic origin. However, research indicates that such compounds feature toxic and carcinogenic effects (Sun-Waterhouse et al. 2011). With the increase of consumer awareness regarding healthy eating, there was a need for natural alternatives, which have the same antioxidant potential, but which do not cause a harmful danger to health.

Studies highlight the presence of phenolic antioxidants and other phytonutrients in oilseeds, important for reducing the incidence of chronic diseases, as long as there is a regular intake of these foods (Melo et al. 2006; Valtueña et al. 2008), with a special interest in the sesame (Sesamum indicum), due to the presence of natural antioxidants, such as phenolic compounds, phytates, lignans and tocopherols (Chen et al. 2005; Kouri and Arriel, 2009).

Sesame ranks ninth among the most cultivated oilseeds in the world. According to the United Nations Organization for Agriculture (Food and agriculture Organization. 2009), sesame is explored in 69 countries, and its world production is estimated at 3.5 million tons of grain, in approximately 7.5 million hectares cultivated. About 90% of the sesame produced
worldwide is destined for food consumption (Kouri and Arriel, 2009), and stands out as a highly nutritious food.

Thus, the objective of this work was to evaluate the oxidative stability of goat hamburgers made with saturated and unsaturated fats under the effect of black sesame extract as a natural antioxidant.

2. Methodology

2.1 Obtaining of extract

Black sesame sample (Sesamum indicum) was acquired from local commerce in the city of Campina Grande - PB, in a dry state. To obtain the extract, two extraction methods were evaluated, being the aqueous method (100% water) and the hydroalcoholic method (70% ethanol and 30% water). Both methods were prepared with 50 g of black sesame and 110 ml of each solvent. After preparation, the extracts underwent a mechanical stirring process for four hours. Then, filtration and drying were carried out, in an air circulation oven at 40°C, for an average time of 18 hours. After drying, the extracts were stored under refrigeration (5 °C) until they were used for the analysis of total phenolic compounds and added to the meat products.

2.2 Assessment of total phenolic compounds of extracts

Evaluation of total phenolic compounds content was determined according to the method of Folin-Ciocalteu (Slinkard and Singleton, 1977). Gallic acid was used for the standard curve and the results were expressed in terms of gallic acid equivalent (mg EAG/g extract). The absorbances of the mixtures were read at 760 nm in UV-vis from Shimadzu, model UV-2550. After analyzing the results, a choice was made between the aqueous and hydroalcoholic extract, which presented the highest content of total phenolic compounds and the highest yield for addition in goat hamburgers.

2.3 Elaboration of goat hamburgers

The meat (85.4%), together with the fat (6%), was subjected to grinding, followed by the addition of refined salt (1.1%), cassava starch (2%), ice water (5%) and finally the
antioxidant (0.5%). Subsequently, the products were hand blended and modeled with the appropriate pieces to shape the hamburger.

Four formulations were made with two repetitions in each treatment. Formulation 1 (F1): with animal fat obtained from bacon and without the addition of antioxidant; Formulation 2 (F2): with animal fat obtained from bacon and addition of 0.5% of black sesame extract; Formulation 3 (F3): with soy oil and without the addition of antioxidant and formulation 4 (F4): with soy oil and the addition of 0.5% black sesame extract. All treatments were stored under freezing at a temperature of -18 ºC until the moment of analysis.

2.4 Physicochemical evaluation of goat hamburgers

The physicochemical analysis were carried out at 0 and 60 days of storage, using the following methodologies:

• Water activity (Aw): performed according to method 978.18, described by AOAC (2000), using an AQUALAB CX2 device (Decagon Devices, Washington, USA);
• Colorimetry: evaluated using the Konica Minolta Colorimeter, model CR-10 for reading the parameters L* (brightness), a* (red/green intensity) and b* (yellow/blue intensity).
• pH: determined using a digital pH meter (DIGIMED, model pH 300M, São Paulo, Brazil), following the parameters described by method no 947.05 from AOAC (2000).
• Water holding capacity (WHC): It was carried out according to the methodology of Grau and Hamm (1953), modified by Hoffmann et al (1982).

2.5 Proximal composition

The moisture, ash and protein contents were determined using the methodology described in items 950.46.41, 920.153 and 928.08, respectively AOAC (2012), and the lipid content was verified following the procedures of Folch, Less and Stanley (1957).

2.6 Oxidation analysis

The lipid oxidation of the products was determined at times 0, 20 and 40 days of storage by the TBARS method, according to the methodology described by Rosmini et al (1996).
2.7 Statistical analysis

All results obtained were submitted to analysis of variance (ANOVA), using completely randomized design, where the statistically different treatments were evaluated by Tukey test at the level of 5% of significance, using the statistical software AGROESTAT.

3. Results and Discussion

The average values of phenolic compounds extracted from black sesame using aqueous and hydroalcoholic solvents are expressed in Table 1.

Table 1 Concentration of total phenolic compounds in black sesame extracts.

| Solvent         | Total phenolic compounds\(^\ast\) (mg EAG / 100 g of sample) | Yield (%) |
|-----------------|---------------------------------------------------------------|-----------|
| Aqueous         | 735.5\(^a\) ± 61.09                                          | 1.0       |
| Hydroalcoholic  | 751.5\(^a\) ± 19.1                                           | 2.4       |

\(^\ast\) Results expressed as mean ± standard deviation (duplicate). Averages followed by the same letter do not differ statistically by the Tukey test (p> 0.05). Source: Authors.

Although no significant difference was observed between the concentration of phenolic compounds in the evaluated extracts, the hydroalcoholic solvent was chosen for application as a natural antioxidant in goat hamburgers because it led to a higher extract yield (2.4%), when compared to the aqueous solvent (1.0%). This fact can be justified due to the use of water in conjunction with ethanol, allowing greater amounts of components to be obtained, since such compounds can present different degrees of polarity, influencing extraction efficiency and yield at the end of the process (Barbi, 2016).

Phenolic compounds play an important role in antioxidant activity through neutralization or sequestration of free radicals and chelation of transition metals, acting both in the initiation stage and in the propagation of the oxidative process (Silva, 2011). The analyzed black sesame extracts presented levels of total phenolic compounds higher than those described by Silva (2011) who reported values of 261.9 ± 7.5 mg in gallic acid equivalent (EAG) per 100 g of seed, indicating that these extracts can be used as ingredients in the preparation of food products, as a strategy to increase the functional properties of food.
Physicochemical characteristics of goat hamburgers are represented in Table 2. There was no significant difference among the formulations when analyzing the moisture content, varying between 69.19 and 69.92%. As it is an abundant component, water influences the quality of the meat, contributing to the juiciness, texture, color and flavor (Lawrie, 2005). On the other hand, water is the main medium for biological reactions that occur in meat, directly affecting such reactions during storage and processing. Santos et al. (2009), working with hamburgers made of different proportions of beef and pork, found similar results for moisture content, which varied from 66.57% to 73.54%, indicating that goat hamburger formulations are similar to the data reported in the literature.

Table 2 Physicochemical characterization of hamburgers made with different lipid sources with or without added antioxidant.

| Parameters | Formulations* |
|------------|---------------|
|            | F1            | F2            | F3            | F4            |
| Moisture (%)| 69.81 ± 0.25  | 69.23 ± 0.48  | 69.92 ± 0.32  | 69.19 ± 0.18  |
| Lipids (%)  | 10.95 ± 0.53  | 11.96 ± 0.98  | 12.88 ± 0.71  | 14.99 ± 1.04  |
| Protein (%) | 13.58 ± 0.14  | 12.51 ± 0.53  | 15.36 ± 0.16  | 11.88 ± 0.53  |
| Ashes (%)   | 1.05 ± 0.1    | 1.38 ± 0.36   | 1.21 ± 0.11   | 1.69 ± 0.05   |
| WHC (%)     | 56.45 ± 2.66  | 59.73 ± 0.39  | 56.45 ± 1.54  | 56.33 ± 2.31  |

* Means followed by equal letters on the same line do not differ statistically by the Tukey test (p > 0.05).
F1- Animal fat without antioxidant; F2 - Animal fat + 0.5% black sesame extract; F3 - soybean oil without antioxidant; F4 - soybean oil + 0.5% black sesame extract. Source: Authors.

Regarding the lipid content, there was a significant difference between the F4 formulation in relation to the F1 and F2 formulations, although it did not differ significantly from the F3 formulation. Lipids are macronutrients of great food importance, present in meat products, in order to improve sensory characteristics, such as softness and juiciness (Gonsalves, 2012). The difference observed among treatments can be justified by the type of fat used, animal fat in formulations F1 and F2 and vegetable soybean oil in F3 and F4. All formulations are in accordance with current legislation, which determines the maximum lipid content corresponding to 23% (Brasil, 2000).

The ash values varied from 1.05 to 1.69% for elaborated goat hamburgers, which shows a statistical difference between the different formulations. These results are close to those reported by Santos et al (2009), which obtained an average ash content of 1.18%. The
ash content is related to the amount of minerals, such as iron, phosphorus, zinc and selenium present in the meat, which provide vital functions for the proper functioning of the central nervous system (CNS), such as controlling muscle contraction, strengthening teeth and bones and transport of oxygen to the blood (Alves, 2005).

Formulation F3 differed significantly from the other formulations in protein content, presenting the highest value of 15.36%. Formulation F1 showed a slightly lower value (13.58%), differing from formulations F2 and F4, with values 12.51 and 11.88%, respectively. The values for the protein contents found are similar to those reported by Martins et al. (2009) of 13.2%, Santos et al (2010) of 13.6% protein and by Lemos et al. (2009) of 15.50%. Protein in meat is considered to be the main responsible for the functional characteristics of emulsion and water holding capacity (WHC), in addition to the nutritional function, considered of high biological value, as it contains all essential amino acids and in proportions that meet the nutritional needs (Pereira, 2013).

Regarding the WHC, there was no significant difference between the formulations, whose results ranged from 56.33 to 59.73%. The water holding capacity (WHC) is of fundamental importance in meat quality during meat storage and/or processing. When the tissues have little water retention capacity, there is loss of moisture and, consequently, weight, also affecting sensory characteristics, such as softness and final yield of the meat product (Goñi, 2010).

Table 3 shows the values of the physicochemical parameters of goat hamburgers evaluated during 60 days of storage.
Table 3 Physicochemical parameters of goat hamburgers during 60 days of storage.

| Parameters       | Formulations | Storage time (days)** |
|------------------|--------------|-----------------------|
|                  |              | 0         | 60         |
| Water activity (Aw) | F1           | 0.99 ± 0.00aA | 0.99 ± 0.00aA |
|                  | F2           | 1.00 ± 0.00aA | 1.00 ± 0.00aA |
|                  | F3           | 1.00 ± 0.00aA | 1.00 ± 0.00aA |
|                  | F4           | 1.00 ± 0.00aA | 1.00 ± 0.00aA |
| L*               | F1           | 42.96 ± 1.27aA | 42.61 ± 1.38aA |
|                  | F2           | 42.43 ± 2.05aA | 42.19 ± 0.68aA |
|                  | F3           | 42.90 ± 1.09aA | 42.48 ± 1.95aA |
|                  | F4           | 41.63 ± 0.92aA | 40.76 ± 2.54aA |
| a*               | F1           | 7.56 ± 0.64abA | 4.96 ± 0.15abB |
|                  | F2           | 7.16 ± 0.80abA | 4.36 ± 0.73abB |
|                  | F3           | 7.60 ± 0.20abA | 4.62 ± 0.71abB |
|                  | F4           | 6.83 ± 0.20abA | 4.36 ± 1.40abB |
| b*               | F1           | 17.16 ± 0.61abA | 18.13 ± 0.15abA |
|                  | F2           | 17.95 ± 0.75abA | 16.26 ± 0.25abB |
|                  | F3           | 17.66 ± 0.45abA | 18.43 ± 0.31abA |
|                  | F4           | 17.13 ± 0.41abA | 16.86 ± 1.22abA |
| pH               | F1           | 5.67 ± 0.04abA | 5.13 ± 0.01abB |
|                  | F2           | 5.40 ± 0.05abA | 5.22 ± 0.02abB |
|                  | F3           | 5.66 ± 0.02abA | 5.11 ± 0.02abB |
|                  | F4           | 5.41 ± 0.03abA | 5.04 ± 0.07abB |

** Means followed by the same letter do not differ statistically from each other by the Tukey test at the level of 5% probability (p >0.05), with lowercase letters for analysis in the columns and uppercase letters for analysis in the lines. F1 - Animal fat without antioxidant; F2 - Animal fat + 0.5% black sesame extract; F3 - soybean oil without antioxidant; F4 - soybean oil + 0.5% black sesame extract. Source: Authors.

Regarding the water activity (Aw), there was no significant difference among formulations at 60 days of storage compared to time 0. Water activity has a fundamental role in the processing to which meat is subjected and mainly in shelf life, as it is a factor favorable to microbial development. The value of Aw gives an indication of the free water content.
present in the food, which is the only form of water used by microorganisms. Bacteria are usually more demanding regarding the availability of free water, followed by molds and yeasts, requiring a high water activity value (Borba, 2010).

Regarding the colorimetric analysis, it was observed that there was no significant difference among all the formulations during the 60 days of storage for parameter L*. Brightness is an important feature for consumer acceptability, as it indicates the freshness of the product at the time of purchase. As for parameter a*, there was a decrease in the value at time 60 for all formulations when compared to time 0. This can be justified by the oxidation of myoglobin to metmyoglobin, thereby reducing the intensity of the red color of the meat. For the parameter b*, a reduction in the yellow color intensity was observed only in the F2 formulation.

When evaluating pH at time 0, higher values were found in F1 and F3 than in F2 and F4. These results may be justified, possibly, by the presence of the natural antioxidant in these latter formulations, which influenced the acidity of goat hamburgers due to the black sesame extract. Within 60 days, formulation F4 presented the lowest pH value, 5.04.

The pH is an important meat quality factor since it is related to the microbial growth in the food, because a drop in the pH value is an indication of lactic acid bacteria growth, while the increase in the pH value is the indication of amine-producing bacteria. Checking the pH over the storage period is a technique to monitor the conservation status of the meat product, and to analyze the growth of some lactic acid bacteria (Morais and Souza, 2012). Therefore, the final pH values ranged from 5.04 to 5.22, which indicates that hamburger formulations are suitable, according to this parameter, for human consumption, as meat products are considered good for consumption up to pH 6.0 (Santos, 2009).

Table 4 shows the lipid oxidation values of goat hamburger formulations over the storage period.
Table 4: TBARs index values expressed in mg of malonaldehyde (MDA)/kg of sample for goat hamburgers over 40 days.

| Formulations | Storage time (days)* |   |   |
|--------------|---------------------|---|---|
|              | 0                   | 20| 40 |
| F1           | 10.63 ± 0.36\textsuperscript{dB} | 11.06 ± 0.39\textsuperscript{cAB} | 11.43 ± 0.07\textsuperscript{cA} |
| F2           | 4.82 ± 0.14\textsuperscript{dA} | 3.33 ± 0.29\textsuperscript{dB} | 3.65 ± 0.08\textsuperscript{dB} |
| F3           | 9.45 ± 0.35\textsuperscript{bB} | 15.91 ± 0.40\textsuperscript{aA} | 18.07 ± 0.18\textsuperscript{aA} |
| F4           | 7.49 ± 0.02\textsuperscript{cC} | 12.12 ± 0.18\textsuperscript{bbB} | 16.03 ± 0.19\textsuperscript{baA} |

* Means followed by the same letter do not differ statistically from each other by the Tukey test at the level of 5% probability (p < 0.05), with lowercase letters for analysis in the columns and uppercase letters for analysis in the lines. F1 - Animal fat without antioxidant; F2 - Animal fat + 0.5% black sesame extract; F3 - soybean oil without antioxidant; F4 - soybean oil + 0.5% black sesame extract. Source: Authors.

Regarding the lipid oxidation values during time 0, it was possible to observe the effect of the formulations with addition of antioxidant (F2 and F4) in relation to their respective formulations without protection (F1 and F3), whose results for oxidation were superior. Formulation F2 showed the lowest lipid oxidation, and did not differ statistically for the values of TBARs between 20 and 40 times, which demonstrates the antioxidant potential of black sesame and less oxidative interferences in this formulation because it is added with saturated animal fat, which has only simple type bonds, being more resistant to undesirable reactions of lipid oxidation.

In formulations without antioxidant, a gradual increase in the lipid oxidation content was observed, as expected by the absence of the antioxidant, especially in the formulation F3 over the days, which in addition to not having protection from free radicals, was formulated with the addition of established fat, whose double bonds are susceptible to the lipid oxidation process (Santos et al. 2013). The low values found in formulation F4 in relation to formulation F3, prove the efficiency of the antioxidant used to delay lipid oxidation in goat hamburgers.

4. Conclusion

Elaborated hamburgers complied with the current legislation regarding the physicochemical parameters, however the type of fat used (saturated and unsaturated) was determinant in the values of malonaldehyde (MDA) observed in the studied goat hamburgers.
The addition of 0.5% of black sesame extract proved to be efficient in controlling oxidation of goat hamburgers, during 40 days of storage, confirming its potential as a natural antioxidant and source of phenolic compounds.

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