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### Physicochemical Characteristics and Lipid Oxidation of Chicken Inner Fillets Subjected to Different Thermal Processing Types

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Cooking, freezing, myofibrillar fragmentation index, lipid oxidation, reheating.

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### **ABSTRACT**

The objective of this study was to evaluate the effects of different types of thermal processing on the physiochemical characteristics and lipid oxidation of chicken inner fillets. The study was divided into three assays. In the first assay, 50 chicken inner fillets were divided into five treatments, totaling 10 samples per treatment. Treatments consisted in cooking in water bath, electric oven, microwave oven, deep frying, or grilling. The analyzed variables were: cooking weight loss (CWL) and lipid oxidation determined by thiobarbituric acid reactive substances (TBARS). In the second assay, 50 chicken inner fillets were divided into five treatments, totaling 10 samples per treatment. Each treatment consisted of the same cooking methods applied in the first assay, and storage for 48 hours under refrigeration and reheating in a microwave oven. The variable analyzed in the second assay was lipid oxidation (TBARS). In the third assay, 30 samples of chicken inner fillets were subjected to one, four and eight freeze-thaw cycles, after which meat pH, myofibrillar fragmentation index (MFI), water retention capacity (WRC), and lipid oxidation (TBARS) were determined. Chicken inner fillets submitted to deep frying and cooked in a microwave oven presented greater lipid oxidation than the other cooking methods, and deep frying resulted in the highest cooking weight loss. Reheating chicken inner fillets in a microwave oven caused the highest meat lipid oxidation. Increasing the number of freeze-thaw cycles increases the pH, MFI, WRC and TBARS values of chicken inner fillets.

#### INTRODUCTION

Brazilian poultry meat market has expanded consistently both domestically and worldwide. Today, Brazil is the leading global chicken meat exporter. However, the competitiveness of this market demands constant technological improvement, standardization, and particularly, strict product quality control (Brossi, 2007).

There is currently a significant demand for boneless chicken cuts, particularly for valuable cuts, such as breast and legs (Pavan *et al.*, 2003). In this scenario, the inner fillet, known sometimes as the fillet mignon of chicken breast, has become popular in the market. This cut is found along the bone and cartilage of the breast, is small and elongated, white in color, low in fat and sold without skin (Lopes Comercial, 2014).

Thermal processing can alter the chemical composition of meat products. Among the different types of processing is cooking, the most commonly applied method for meat consumption. Cooking promotes the Maillard reaction, which is a conversion of collagen into gelatin, protein denaturation and can lead to changes in fats, resulting in a peculiar odor in each meat type (Bobbio & Bobbio, 1995).



Reheating is another heating method that can alter the chemical composition of pre-cooked meats due to the possible oxidation of polyunsaturated fatty acids, resulting in the development of warmed over flavor (WOF) (Pearson et al., 1977; Willemot et al., 1985). The development of undesirable aromas or flavors is a great challenge for meat processors due to the increasing demand for pre-cooked meats in recent decades (Luciano et al., 2009).

Another type of thermal processing that can cause positive and negative effects in meat products is freezing, which is used to preserve meat quality, both domestically and industrially. However, gradual losses in quality attributes may occur during storage under low temperatures, such as oxidation, osmotic release of water, and protein denaturation, adversely affecting the shelf life of frozen products (Benjakul *et al.*, 2003). Among these changes, lipid oxidation is considered the main factor responsible for flavor deterioration and reduced shelf life of products due to the onset of peroxidation (Angelo *et al.*, 2009).

Therefore, this study was conducted to evaluate the effects of different types of thermal processing on the physiochemical characteristics and lipid oxidation of chicken inner fillets.

### **MATERIAL AND METHODS**

Three assays were performed between February and July 2014. Chilled chicken inner breast fillets from the same batch were obtained from a broiler processing plant, where inner fillets were placed in plastic containers (five per container), labeled, and frozen at -18°C for 30 days until analyses. The three assays were conducted with different samples.

### Assay I

In the first assay, 50 inner fillets were distributed in a completely randomized experimental design, with five treatments of 10 replicates each. Each treatment consisted of a cooking method and each inner fillet was regarded as one experimental unit.

The previously frozen samples were thawed, weighed (initial weight), and subjected to lipid oxidation analysis by the thiobarbituric acid reactive substances method (TBARS), according to the methodology described by Vyncke (1970).

For thawing, samples were arranged on a polyethylene tray, protected by a plastic film and kept in under refrigeration (6°C) for 12 hours; therefore, all inner fillets were submitted to the same thawing conditions.

Samples were then individually submitted to one of following cooking methods:

- a) Water bath (WB): samples were placed in polyethylene bags, and immersed in water preheated to 80°C for approximately six minutes until reaching 72°C internal temperature.
- b) Electric oven (EO): samples were placed in a non-stick aluminum baking pan, and baked in an electric oven preheated to 150 °C for approximately six minutes until reaching 72°C internal temperature.
- c) Microwave oven (MO): samples were placed in a glass bowl and cooked at maximum power for approximately two and a half minutes, until reaching 72°C internal temperature.
- d) Deep frying (DF): samples were placed in a nonstick frying pan with 15 mL hot soybean oil (± 160°C), and turned once after one and a half minute, totaling three minutes of oil immersion, until reaching 72°C internal temperature. After frying, samples were placed on absorbent paper to remove oil excess and to cool down.
- e) Grilling (GR): samples were placed on an electric grill preheated to 180°C for approximately eight minutes; each chicken inner fillet was turned once after approximately four minutes until reaching 72°C internal temperature.

The internal temperature of the individual samples was determined using a meat thermometer. The time and temperature used in each cooking method were determined in a previous assay.

After cooking, the samples were left to reach room temperature, placed on polyethylene trays and wrapped in plastic film, weighed (final weight), and again analyzed for lipid oxidation (mg of malonaldehyde per kg of sample) using TBARS. In order to correct for the lipid oxidation values measured in the fresh samples, which might have been different at the time of commercial acquisition), lipid oxidation values were determined as the difference between malondialdehyde values obtained before and after cooking. Cooking weight loss (CWL) was calculated as the difference between initial and final weights.

#### **Assay II**

In the second assay, 50 inner fillets were distributed according to a completely randomized experimental design into five treatments of 10 replicates each. Each chicken inner fillet was considered as one experimental unit. After freezing, samples were thawed and submitted to the same cooking methods as described



in the first assay, cooled to room temperature, and subjected to lipid oxidation analysis (Vyncke, 1970). Samples were then placed on polyethylene trays, wrapped in plastic film, and stored under refrigeration (6°C) for 48 hours, after which they were reheated for four minutes in a microwave oven at high power, cooled to room temperature, and again analyzed for lipid oxidation (Vyncke, 1970).

Lipid oxidation was calculated as the difference between malondialdehyde values obtained in the cooked samples in assay I and after reheating in the microwave oven.

### **Assay III**

In the third assay, 30 inner fillets were distributed according to a completely randomized design experimental into three treatments (freezing/thawing cycles) with 30 replicates of one chicken inner fillet each (experimental unit). Samples were wrapped, individually numbered, and subjected to three different freeze/thaw cycles, i.e., samples were frozen and thawed once, four, or eight times, as described below.

Firstly, samples were individually weighed in an analytical balance (± 0.0001g) and longitudinally cut into three equal sections, each of which was submitted to a freeze/thaw cycle, and the remaining parts were returned to the freezer (-18°C). After freezing, the samples were removed from the freezer in the afternoon and remained under refrigeration (6°C) overnight until completely thawed. The next morning, the part to be analyzed was removed from the same samples and the other two were returned to the freezer for the second freezing cycle. The same procedure was adopted for the samples submitted to four and eight cycles.

The following parameters were analyzed in the samples submitted to the freeze/thaw cycles: pH, myofibrillar fragmentation index (MFI) (Culler *et al.*, 2009), water retention capacity (WRC) (Lakshmanan *et al.*, 2007), and lipid oxidation (TBARS) (Vyncke, 1970).

Data were analyzed for normality using the Shapiro-Wilk test, and then submitted to analysis of variance. Means were compared by Tukey's test ( $p \le 0.05$ ). All statistical analyses were carried out using SAS statistical software.

### **RESULTS AND DISCUSSION**

The different cooking methods evaluated influenced (p<0.05) of cooking weight loss (Table 1) of the broiler inner fillets, with the lowest values observed by cooking in water bath (16.70%) and in the microwave oven (17.24%).

**Table 1** – Initial weight (IW), final weight (FW), and mean cooking weight loss (CWL) values of chicken inner fillets subjected to different cooking methods.

|                | IW (g) | FW (g) | CWL (%)             |
|----------------|--------|--------|---------------------|
| Water Bath     | 33.26  | 27.68  | 16.70°              |
| Electric Oven  | 34.45  | 26.44  | 23.48 <sup>b</sup>  |
| Microwave Oven | 30.71  | 25.56  | 17.24 <sup>c</sup>  |
| Deep Frying    | 34.21  | 24.07  | 30.11ª              |
| Grilling       | 37.30  | 30.15  | 19.39 <sup>bc</sup> |
| Mean           | -      | -      | 21.38               |
| SEM            | -      | -      | 0.88                |

<sup>\*</sup>Standard Error of the Mean. Means in the same row, followed by different lowercase letters, differ significantly by Tukey's test ( $p \le 0.05$ ).

When using conventional heating forms (direct flame, hot air, direct contact with hot grill, and others) for cooking meat, heat sources cause the food molecules to heat up from the surface to the inside of the muscle mass, so that heating occurs in successive layers. Therefore, the outside layers of the cut are cooked first, i.e., the protein is coagulated, forming a film, which prevents the loss of meat components to the outside before its internal temperature rises, resulting in lower cooking loss (Potter & Hotchkiss, 1995). On the other hand, in a microwave oven, heat is transferred by electromagnetic irradiation emitted by a warm body and absorbed by a cold body, increasing kinetic energy and causing thermal excitation. This promotes homogenous temperature distribution from the area where the temperature is high to where the temperature is low (Araújo, 1982). When food is cooked in a microwave oven, heat is quickly generated and evenly distributed throughout the food item, as the inner molecules boils and the generated steam heats the adjacent solids by conduction and is lost to the outside (Girard, 1991), causing greater water loss compared with direct heating methods. However, in the present study, as soon as the internal temperature of the meat reached 72 °C, heating was interrupted, minimizing water loss to the outside.

The results of the present study are partially different from those obtained by Rosa *et al.* (2006), who compared different cooking methods (immersion in water, conventional oven, grill, microwave oven, and deep frying) of chicken breast and leg, and observed greater cooking weight loss with a microwave oven (32.49%), followed by deep frying (29.18%), and the lowest cooking weight loss (CWL) when samples were cooked in a grill (23.46%).

The higher cooking weight loss found by Rosa *et al.* (2006) using a microwave may be explained by cooking time, which was 10 minutes for breast samples and 12 minutes for leg samples – longer than that used in



this study. In addition, those authors do not report the internal temperature measurements, which might have been higher than 72°C, leading to greater evaporation of meat components. Moreover, inner fillets are cut differently from conventional chicken carcass parts, which may also have influenced the result.

The results of the present study partially corroborate the findings of Vieira (2005), who compared chicken fillets cooked in water, deep fried, and roasted in a conventional oven or in a microwave oven, and reported lower cooking weight losses in deep-fried and water-cooked samples.

According to Califano et al. (1997), in meat products heated between 40°C and 60°C, there is considerable actin denaturation, and this effect is responsible for the loss of fluids that occurs in the product. However, literature defines meat cooked at internal temperatures of 60 °C as "rare", 71 °C "medium" and 77 °C "well done" (Savell et al., 1998), indicating greater water losses when meat is cooked at higher temperatures. The highest cooking loss obtained with the deep frying method in the present study may be explained by the high temperature to which the samples were exposed to, leading to greater protein denaturation and, consequently, greater meat fluid loss.

The greater differences in malonaldehyde concentrations between fresh and cooked chicken inner were obtained with the deep frying and microwave oven methods (p<0.05). The lowest difference was calculated when samples were grilled (Table 2), whereas the values of samples cooked in water bath and in electric oven were not different (p>0.05) from the other treatments.

**Table 2 –** Values of the differences in malonaldehyde concentrations of chicken inner fillets of assay I (fresh and cooked) and assay II (cooked and reheated)

|                | Malonaldehyde (mg/kg) |                      |  |
|----------------|-----------------------|----------------------|--|
|                | Difference*           | Difference**         |  |
| Water Bath     | 0.1621 <sup>ab</sup>  | 0.2222b              |  |
| Electric Oven  | 0.1293 <sup>ab</sup>  | 0.0763b              |  |
| Microwave Oven | 0.1783ª               | 0.6533ª              |  |
| Deep Frying    | 0.1871ª               | -0.0291 <sup>b</sup> |  |
| Grilling       | 0.0989 <sup>b</sup>   | 0.0570 <sup>b</sup>  |  |
| Mean           | 0.1511                | 0.1959               |  |
| SEM***         | 0.0080                | 0.0451               |  |
| p Value        | 0.0022                | <0.0001              |  |

<sup>\*</sup>Assay I - Difference: cooked - fresh \*\* Assay II - Difference: reheated - cooked

Means in the same column followed by different lowercase letters significantly differ by Tukey's test ( $p \le 0.05$ ).

The microwave oven cooking method caused greater oxidation possibly because it is directly linked

to reactions of fat oxidation initiation. Lipid oxidation consists of three main phases: initiation, propagation, and termination (Sevanian & Hochstein, 1985), and in meats, fat oxidation initiation reactions can take place through the actual thermal degradation of organic matter or by using microwave ovens by energy absorption (Ferrari, 1998).

Deep frying also promoted high lipid oxidation differences between fresh and cooked inner fillets, and may be explained by actual autoxidation, which is the main mechanism of oxidation of oils and fats (Berger & Hamilton, 1995). Therefore, immersing meat samples in oil likely increases lipid levels in their composition, causing greater lipid oxidation in the samples subjected to the deep frying process. In order to prevent oil autoxidation, the incidence of all factors that favor it must be reduced, lowering as much as possible the levels of temperature and light (Ramalho & Jorge, 2006). However, it is essential to increase the temperature when oils are used for cooking, which may add up to the change in lipid oxidation when foods are deep fried, as it combines the high temperature of the cooking process with the greater availability of fatty acids susceptible to oxidation (Silva, 2013).

The oxidation processes during cooking are more influenced by long cooking time and lower temperature than by short cooking time and higher temperature (Broncano et al., 2009). Because the samples cooked in the microwave oven presented higher levels of oxidation compounds, despite the low temperature and short time, it is suggested that there may be an effect of the microwave oven on meat fat that causes oxidation of polyunsaturated fatty acids (Broncano et al., 2009), which may explain the increase in lipid oxidation observed in the microwave oven cooking process. Broncano et al. (2009), analyzing the Latissimus dorsi muscle of pigs treated with different cooking methods, determined that cooking in microwave oven caused slightly lower lipid oxidation compared with frying and roasting, as partially observed in the present study.

Some enzymes can inhibit lipid oxidation, such as glutathione peroxidase, which catalyzes the reduction of organic hydroperoxides, protecting lipoproteins and cell membranes. However, the activity of glutathione peroxidase, as well as that of most enzymes, is reduced as heating temperature increases, which may lead to greater lipid oxidation of food items, regardless of cooking method (Thomas, 1990; Arthur, 2000), as detected by the current results.

The differences in malonaldehyde concentrations between the cooked and reheated samples was greater

<sup>\*\*\*</sup>Standard Error of the Mean.



(p<0.05) in inner fillets cooked in the microwave oven compared to the other treatments (Table 2).

TBARS values greater than 1 mg of malonaldehyde per kg of sample are perceptible, as they cause off flavors (Djenane *et al.*, 2002). The samples that showed values near 1 mg were those cooked and reheated in the microwave oven. The results showed that cooking in a microwave oven was the process that most influenced (60.21%) lipid oxidation when the chicken inner fillets were reheated, compared to the fresh samples.

Cooking is one of the main causes of lipid oxidation of meat products, and protein denaturation occurs during this process due to heating. However, when reheating pre-cooked meats, the chelation of denaturated proteins releases active iron, which acts as a catalyzer of oxidation reactions that are responsible for the development of warmed over flavor in reheated meats (Igene *et al.*, 1980).

The different freeze/thaw cycles (Table 3) influenced (p<0.05) meat pH values. The pH values obtained after the first cycle were lower (6.03) compared with to the other freezing cycles. On the other hand, from the fourth freezing cycle, pH values remained unchanged, indicating pH stabilization.

**Table 3** – Mean pH, malonaldehyde concentration, myofibrillar fragmentation index (MFI) and water retention capacity (WRC) values of chicken inner fillets subjected to different freeze/thaw cycles

| Freeze/thaw cycles | рН                | Malonaldehyde<br>(mg/kg) | MFI (%)            | WRC (%)            |
|--------------------|-------------------|--------------------------|--------------------|--------------------|
| 1 cycle            | 6.03 <sup>b</sup> | 0.1439 <sup>c</sup>      | 58.51 <sup>c</sup> | 63.06°             |
| 4 cycles           | 6.22ª             | 0.4035 <sup>b</sup>      | 63.57⁵             | 64.10 <sup>b</sup> |
| 8 cycles           | 6.25ª             | $0.4909^{a}$             | 67.65ª             | 65.21ª             |
| Mean               | 6.17              | 0.3461                   | 63.24              | 64.13              |
| SEM*               | 0.02              | 0.0159                   | 0.47               | 0.19               |
| p Value            | <0.0001           | <0.0001                  | <0.0001            | <0.0001            |

<sup>\*</sup> Standard Error of the Mean. Means in the same column, followed by different lower-case letters, differ significantly by Tukey's test (p $\leq$ 0.05).

These results are different from those obtained by Ali *et al.* (2015), who analyzed the influence of freezing cycles (six cycles) on the pH of chicken breast fillets and observed that pH decreased as freezing cycles increased.

Freeze/thaw cycles significantly affected water retention capacity (WRC). The highest values were found in the inner fillets subjected to eight freeze/thaw cycles, and the lowest values in those frozen and thawed only once. The inner fillets that were frozen four times differed from the remaining treatments. When studying the effect of freezing on chicken meat, Hernandez (2005) obtained lower WRC values in fresh

meat. It is known that WRC is influenced by general factors, among which the most important are pH and myofibrillar fragmentation index (MFI) (Forrest *et al.*, 1979), and it is defined as the capacity of meat to retain its own water during the application of external forces, such as freezing. Consequently, it is assumed that the stronger the external forces applied on the meat, the lower is its water retention capacity.

Lower WRC and pH values when determined in the inner fillets subjected to eight freezing cycles, confirming the positive correlation between these variables (Ngapo et al., 1999; Zapata et al., 2006) and suggesting that higher pH values increase the capacity of the meat to retain water.

The myofibrillar fragmentation index (MFI) is frequently used in meat quality assessments, as it predicts more than 50% of the variation in meat tenderness (Hopkins *et al.*, 2000; Veiseth *et al.*, 2001). In the present study, the samples were not large enough to determine shearing force or sensorial characteristics. MFI values were different (*p*<0.05) among the evaluated freezing cycles, and increased as the number of freeze/thaw cycles increased. According to Culler *et al.* (2009), meats with MFI values higher than 60 are considered to have satisfactory texture, and overall, the MFI values determined in the present study were close the recommended value (average 63.24).

The fragmentation of myofibrils caused by autolysis and the severity of the cold/heat cycles may have influenced the increase in WRC, given that the pH did not show significant differences between treatments following thawing/freezing cycles.

Malonaldehyde level increased (*p*<0.05) as the number of freeze/thaw cycles increased, with the highest values of malonaldehyde found when subjected to eight freezing cycles, followed by four, and one freeze/thaw cycle. These results are consistent with those of Ali *et al.* (2015), who studied the effect of freeze/thaw cycles (six cycles) on lipid oxidation and observed a considerable increase in the concentration of malonaldehyde in thawed chicken breasts after the fourth freeze/thaw cycle, and even greater concentrations after the fifth and sixth cycles.

Research on cooking methods and freezing cycles of meat products are still scarce, which emphasizes the importance of studies evaluating the effect of thermal processing methods on meat products in order to disseminate that knowledge to the public, as well as to reduce the consumption of oxidized food items by consumers, considering that meat cooking and freezing are also household processes.



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### **CONCLUSION**

Deep frying and cooking in a microwave oven are the cooking methods that cause the highest lipid oxidation in chicken inner fillets. Cooking weight loss is the highest when the fillets are deep fried.

When reheating chicken inner breast fillets, the use of microwave oven increases lipid oxidation.

The three freeze/thaw cycles (one, four and eight cycles) increase meat pH, water retention capacity, myofibrillar fragmentation index, and lipid oxidation, impairing the quality of broiler chicken inner fillets.

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