

# Evaluation of indicative parameters of the quality of ground beef sold in different supermarkets of Erechim, Rio Grande do Sul

## Avaliação de parâmetros indicativos da qualidade da carne bovina moída comercializada em diferentes supermercados em Erechim, Rio Grande do Sul

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### ABSTRACT

**Introduction:** In the last decades, Brazilians, following a worldwide trend, have been changing their eating habits, worrying about the origin of food. **Objectives:** The objective of this study was to evaluate the physicochemical and microbiological parameters of ground beef sold in different supermarkets in the city of Erechim, RS. **Method:** Three collections were performed in four different establishments, totalizing twelve samples. The meat quality was evaluated through the following parameters temperature, pH, determination of molds and yeasts, proof of cooking, of Nessler and Éber, search of nitrate, nitrite and sodium sulfite. **Results:** From the results it was possible to verify that all the samples collected did not comply with the legislation in relation to storage temperature, and presented high count of molds and yeasts. However, all samples were within the established standard for pH values. Nonetheless, unpleasant odors were present in 25.0% of the samples, within positive results for the Nessler and Éber tests, and negative for 100.0% of the samples in relation to fraud determinations. **Conclusions:** From these results, becomes evident the need for actions that aim to obtain and commercialize higher quality ground beef in the city Erechim. Therefore, it is important to improve the sanitary education and the awareness of entrepreneurs and employees who work in the sector, also more active inspection of the Sanitary Surveillance of the Municipality.

**KEYWORDS:** Physical-chemical Analysis; Microbiological Analysis; Fraud; Sanitary Surveillance

### RESUMO

**Introdução:** Nas últimas décadas, os brasileiros, seguindo uma tendência mundial, vêm mudando os seus hábitos alimentares, preocupando-se com a origem dos alimentos. **Objetivo:** O objetivo deste estudo foi avaliar os parâmetros físico-químicos e microbiológicos da carne bovina moída, comercializada em diferentes supermercados no município de Erechim, Rio Grande do Sul. **Método:** Foram realizadas três coletas, em quatro estabelecimentos diferentes, totalizando 12 amostras. A qualidade da carne foi avaliada através dos parâmetros temperatura, pH, determinação de bolores e leveduras, prova de cocção, de Nessler e de Éber; pesquisa de nitrato, nitrito e sulfito de sódio. **Resultados:** A partir dos resultados foi possível verificar que todas as amostras coletadas não atenderam a legislação em relação à temperatura de armazenamento, e apresentaram uma elevada contagem de bolores e leveduras. No entanto, todas as amostras encontraram-se dentro do padrão estabelecido para os valores de pH. Verificou-se a presença de odores desagradáveis em 25,0% das amostras, resultados positivos para a prova de Nessler e Éber, e negativos para 100,0% das amostras em relação as determinações de fraudes. **Conclusões:** A partir destes resultados, fica evidente a necessidade de condutas que visam a obtenção e a comercialização de carne moída com maior qualidade no município de Erechim. Por isso, é importante melhorar a educação sanitária e a conscientização dos empresários e colaboradores que atuam no setor, além da fiscalização mais ativa da Vigilância Sanitária do Município.

**PALAVRAS-CHAVE:** Análises Físico-químicas; Análise Microbiológica; Fraude; Vigilância Sanitária

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Received: Apr 10, 2017

Approved: Oct 17, 2017



## INTRODUCTION

Following a global trend, Brazilians have changed their eating habits over the last decades. Brazilian consumers now worry about food origin, quality and possible health risks related to food consumption. Aspects that were previously overlooked like food safety, hygiene, quality and reliability have become important factors for decision making at the time of purchase.

Therefore, for the production of safe food, in addition to considering the consumers' demands, we need to involve all the professionals working in product processing. Government agencies must also adopt effective measures concerning the inspection of the food production chain.

Among meat products, ground beef stands out because it is well accepted by consumers: it is easy to prepare, versatile and fairly inexpensive<sup>1</sup>. However, its quality is a global concern because it has a larger contact surface, it is more frequently handled and often has poor storage temperature control, which favors the growth of microorganisms and oxidation reactions<sup>2,3</sup>.

Additionally, in some commercial establishments, tools used to make ground beef, such as meat grinders, knives and inventory materials, are often not regularly sanitized, which increases the possibility of contamination<sup>4</sup>.

Ground beef is a product that quickly acquires unpleasant sensory characteristics, therefore, to reduce losses with its deterioration, some merchants adopt fraudulent strategies, like the addition of nitrate, nitrite and sodium sulfite. However, the addition of these preservatives is not allowed in fresh meat<sup>5</sup> because this illegal practice may cause adverse health effects to consumers due to the toxicity of some additives.

Because of the characteristics of ground beef and possible fraudulent actions, some cities in Brazil - like Recife - have banned the marketing of pre-ground beef in supermarkets, grocery stores and similar establishments, according to Law n. 17.721, of June 29, 2011<sup>6</sup>. The law rules that meat can only be ground in the presence of the consumer in order to enable greater quality control and avoid contamination.

The use of meats unfit for consumption and the use of illegal substances to conceal their sensory characteristics gave rise to the Weak Flesh Operation, set off by the Brazilian Federal Police in March 2017, after irregularity reports in 21 meatpacking plants.

The discovery of irregularities in meat products is generating great apprehension among consumers in the Brazilian state of Rio Grande do Sul (RS). The state accounts for roughly 8% of Brazil's beef production and for the exports of 124 thousand tons of meat products in 2016, i.e. 8.8% of the country's exports of beef and beef-based products<sup>7</sup>.

Therefore, ground beef is a product that needs more attention, especially in relation to microbial growth and fraud. It is important to conduct research to assess ground beef quality in order to improve public health. Thus, considering the relevance of the

topic and the lack of studies on the quality of the meat marketed in the city of Erechim, Brazil, the objective of this study was to evaluate the physico-chemical and microbiological parameters of the ground beef sold at different supermarkets in the aforementioned city.

## METHOD

### Sample collection and preparation

The research adopted an exploratory descriptive approach, characterized as a case study. From September to November 2016, three samples of ground beef were collected. They were purchased in four different marketplaces, totaling 12 samples.

Samples of approximately 500 g were collected at each establishment, randomly selected, in the city of Erechim, located in the state of Rio Grande do Sul, Brazil.

At the supermarkets, the beef was packed in polyvinyl chloride (PVC) plastic film and stored in refrigerated counters, ready to be marketed. At the time of collection, ground beef temperature was measured using a BT TIP 439 infrared thermometer. The samples were then placed in an ice-filled isothermal box and immediately sent to the laboratory of the Federal Institute of Education, Science and Technology of Rio Grande do Sul, for physico-chemical analyses, qualitative analytical determinations of fraud and microbiological analysis, which were done in triplicates.

### Physico-chemical analytical determinations

The evaluation of the physico-chemical characteristics of pH, cooking test, Eber's test, Nessler's test, nitrate, nitrite and sodium sulfite test followed the standards recommended by the Brazilian Ministry of Agriculture, Livestock Farming and Food Supply (MAPA)<sup>8</sup> and the analytical standards of the Adolfo Lutz Institute<sup>9</sup>.

### pH determination

The pH was determined by the potentiometric method. For this, 50 g of each sample were mixed with 10 mL of distilled water and the pH was measured in an mPA-210 Tecnopon digital potentiometer.

### Cooking test

We placed 20 g of beef in a 250 mL beaker and filled it with distilled water until the sample was covered. The beaker was covered with a watch glass and then heated to 100° C in water bath to start the first vapors so we could evaluate the odors produced. The perception of ammonia or hydrogen sulfide odors evidence deteriorating beef.

### Eber's test - reaction for hydrogen sulfide gas

We performed Eber's test for hydrogen sulfide gas (H<sub>2</sub>S) by transferring 10 g of the homogenized sample to a 125 mL titration flask, which was closed with two overlapping paper filter discs.



The paper surface was soaked with lead acetate solution, and thereafter the flask was placed in water bath at 100 ° C for 10 min. Black spots on the filter paper due to contact with the vapors would indicate the presence of H<sub>2</sub>S gas. The negative reaction would be indicated by the production of spots less intense than those produced by 0.1 mg of Na<sub>2</sub>S<sub>9</sub>H<sub>2</sub>O in an acid medium, which corresponds to 0.014 mg of H<sub>2</sub>S under the conditions of the method we adopted.

#### Nessler's test

For Nessler's test, 10 g of the sample were placed in a titration flask with ground stopper. We then added 100 mL of distilled water to it. After this step, stirring was performed for 15 min with 2 min rest intervals. The solution was filtered with Whatman #1 filter paper. 10 mL of the filtrate were then transferred to a test tube into which Nessler's reagent was added. It was considered a positive result when the filtrate presented a yellow to orange color and a negative result when the color was greenish yellow.

#### Qualitative analytical determination of nitrate and nitrite

To assess the presence of nitrate and sodium nitrite, the samples were first clarified from 10 g of beef, which were placed in a 150 mL beaker with 60 mL of hot distilled water. The beaker was placed in a 60 ° C water bath, and the solution was constantly mixed for one hour. The solution was filtered, cooled to room temperature and used for nitrate and sodium nitrite assessment.

1 mL of the filtrate, some crystals of sodium azide and two drops of sulfuric acid were placed in a test tube. We let the solution stand for five minutes and then heated it to boiling. We then cooled it to room temperature, and a drop of saturated sodium chloride solution and 4 mL of diphenylamine was added. The samples were kept at rest for one hour before we assessed the results. Blue spots would indicate the presence of nitrate in the sample.

Nitrite assessment was performed by adding 10 mL of the filtrate, 1 mL of sulfanilic acid and 1 mL of alpha naphthylamine in a test tube. The tube was shaken and held for 30 min. The pink color would indicate the presence of nitrite in the sample.

#### Qualitative analytical determination of sodium sulfite

3.5 g of the sample were placed in a porcelain dish and 0.5 mL of 0.02% malachite green solution was added to it. With a spatula, we blended the sample for 2 min. In the presence of sulfite, the solution would become colorless.

#### Mold and yeast counts

The counting was performed by weighing 25 g of each sample in a sterile plastic bag in a Stomacher® blender, followed by the transfer of 225 mL of peptone saline diluent at 0.10%. We thus obtained a 10<sup>-1</sup> dilution. After homogenization of the sample, serial dilutions were carried out up to 10<sup>-3</sup>. For the sowing process, we used the surface plating technique through acidified PDA (Potato Dextrose Agar) with 10% tartaric acid, at a pH of 3.5. Plates were then incubated at 25 ± 1 ° C for a period of 5 days.

After this procedure, mold and yeast colonies were counted in colony counters (CP600Plus, Phoenix®) and the results were expressed by the number of Colony Forming Units per gram of sample (CFU.g-1)<sup>10</sup>.

#### Statistical analysis

The experiment was carried out based on a completely randomized design. The samples were collected from a 4 x 3 factorial arrangement (four supermarkets in three collections). Experimental sampling was performed in each sample, totaling 12 samples, evaluated in triplicates. For analysis of the studied parameters, the Statistical 8.0 software was used in the Univariate Analysis of Variance module (*One-way ANOVA*), and the Tukey test at the 5% probability level ( $p < 0.05$ ) for comparison of means. Other statistical data was generated in the *Basic Statistics* module.

## RESULTS AND DISCUSSION

The mean values obtained for the temperature (Table 1) of the ground beef sold at the surveyed supermarkets showed that none of the samples complied with Regulatory Instruction n. 83, of November 21, 2003, of the MAPA<sup>5</sup>, which rules that chilled ground beef must be kept at a temperature of 0 ° C to 4 ° C. The samples collected in Supermarket B had a lower average value (5.98 ° C), differing ( $p < 0.05$ ) from the values obtained in the samples collected in the other supermarkets. Similar values were found by Arçari et al.<sup>11</sup>, who analyzed 25 samples of ground beef from five different supermarkets in the city of Vitória, Espírito Santo, Brazil, and found that only one supermarket (five samples) was in compliance with the standards of the legislation in force.

Baptista et al.<sup>12</sup>, when studying quality aspects of ground beef marketed in the metropolitan area of Recife, Pernambuco, Brazil, found that only two of the 20 meat samples collected in different establishments had internal temperature below 4 ° C. These authors pointed out that the lack of proper ground beef conservation poses a great risk to the health of the consumers. As we said before, because of the greater contact surface, ground beef is more exposed to contamination.

However, it should be noted that some municipal and state laws rule that chilled ground beef may be kept at a temperature below 7 ° C, a value found only in the samples collected at Supermarket B. Matos et al.<sup>13</sup> studied the health profile of beef marketed in different supermarkets in the city of Santo Antônio de Jesus, Bahia, Brazil, and found that 55% of the samples ( $n = 20$ ) had temperatures above 7 ° C.

According to Ritter et al.<sup>14</sup>, several factors influence the microbial population found in the meat, among them the storage temperature in points of sale and retail. Therefore, it is essential that consumers are demanding at the time of purchase. Furthermore, all professionals involved in the activities related to product processing and government agencies should take effective surveillance measures to preserve the health of the population.



**Table 1.** Results of the mean  $\pm$  standard deviation of temperature, pH and mold and yeast counts obtained after the analysis of the samples collected in different supermarkets.

Supermarket	Temperature ( $^{\circ}$ C)	pH	*Mold and yeast
A	9.48 $\pm$ 0.6 <sup>a</sup>	5.82 $\pm$ 0.14 <sup>a</sup>	3.9 $\pm$ 0.29 <sup>a</sup>
B	5.98 $\pm$ 1.6 <sup>b</sup>	5.64 $\pm$ 0.19 <sup>a</sup>	3.2 $\pm$ 1.58 <sup>a</sup>
C	8.46 $\pm$ 2.1 <sup>a</sup>	5.56 $\pm$ 0.27 <sup>a</sup>	2.8 $\pm$ 1.40 <sup>a</sup>
D	8.63 $\pm$ 0.3 <sup>a</sup>	5.67 $\pm$ 0.26 <sup>a</sup>	4.1 $\pm$ 1.18 <sup>a</sup>

\*Mold and yeast count expressed through the decimal logarithm of the number of colonies ( $\text{Log}^{\text{CFU.g}^{-1}}$ ). Different letters in the same column differ from each other ( $p < 0.05$ ) by the Tukey test;  $n = 12$  (36 replicates).

Regarding pH values, all analyzed samples were within the standard established by the MAPA<sup>8</sup>, with mean pH values lower than 6.2, indicating that the beef was fit for consumption.

Similar results for pH values were also observed by Velho et al.<sup>15</sup>, when analyzing 48 samples of fresh beef marketed in the city of Mossoró, Rio Grande do Norte, Brazil. The authors observed that the analyzed samples had pH values ranging from 5.45 to 5.73.

Marchi et al.<sup>16</sup> analyzed the physico-chemical and microbiological parameters of 30 ground beef samples collected at different supermarkets and butchers in the city of Jaboticabal, São Paulo, Brazil, and found that 40% of the samples presented higher pH values than those recommended by the MAPA<sup>5</sup>. While Souza et al.<sup>17</sup> studied the microbiological and physico-chemical quality of 30 samples of ground beef marketed in butchers in the city of Macapá, Amapá, Brazil, and verified that all complied with the legislation regarding pH, but they had high populations of thermotolerant coliforms and *Staphylococcus aureus*. Skrócki<sup>18</sup> found that the analyzed beef samples had populations of aerobic microorganisms, whereas the pH values were between 5.5 and 6.2.

Similar results were obtained in this study because, although pH values were lower than 6.2, the samples collected in the four supermarkets showed mold and yeast counts (Table 1), with highlights to the samples collected in Supermarket D, which presented a mean value of 4.1  $\text{LogCFU.g}^{-1}$ .

The mean values obtained in this study were higher than the results found by Rossi Jr. et al.<sup>19</sup>, who found a mold and yeast count of  $5.9 \times 10^2 \text{ CFU.g}^{-1}$  (2.7  $\text{LogCFU.g}^{-1}$ ) for manual deboning on the counter and  $6.2 \times 10^2 \text{ CFU.g}^{-1}$  (2.8  $\text{LogCFU.g}^{-1}$ ) for air manual boning in mechanically separated beef. Like Hoffmann et al.<sup>20</sup>, who obtained counts of  $7.0 \times 10^1$  to  $2.4 \times 10^2 \text{ CFU.g}^{-1}$  in ground beef, values corresponding to 1.8 and 2.4 log units.

High mold and yeast values indicate poor hygienic-sanitary conditions in equipment and tools, contaminated raw material, and process or storage failure<sup>21</sup>. Veld.<sup>22</sup> and Marchi, et al.<sup>16</sup> also found high values for mold and yeast counts, which ranged from  $10^2$  to  $10^6 \text{ CFU.g}^{-1}$ , corresponding to 2 and 6  $\text{LogCFU.g}^{-1}$ .

It should be noted that Brazilian legislation does not establish limits for molds and yeasts, but this is a group of microorganisms that can produce mycotoxins and accelerate beef decay. The high counts of these microorganisms indicate the need for adequate sanitizing methods in the establishments we studied<sup>23</sup>. Mold and yeast growth is also favored by the use of wooden tools, which

absorb moisture and become impregnated with organic matter, making them ideal spots for the proliferation of microorganisms<sup>24</sup>.

Therefore, according to Kochanski et al.<sup>25</sup>, the proper hygiene of equipment, tools and professionals is an important factor for the control of product quality.

As a result of the increased demand of consumers, competitive pressure and frequent changes in markets and technologies, organizations are urged to constantly improve their products and processes<sup>26,27</sup>. In this context, the concept of "quality management system" stands out. That is a tool created and used with the purpose of offering consumers safe and quality products. Therefore, the integration of quality tools is fundamental to food safety. These tools are to be applied throughout the chain, from the production to the consumers, enhancing the communication of food distributors and regulatory authorities<sup>28</sup>.

The absence of specific legislation in Brazil and the pursuit of quality control have forced some Brazilian states to define their own standards. That is the case of the state of São Paulo, which establishes standards for molds and yeasts in fresh meat of at most  $10^3 \text{ CFU.g}^{-1}$ , i.e. 3  $\text{LogCFU.g}^{-1}$ <sup>29</sup>. Considering this value as a standard, we observed that the samples analyzed in this study may represent public health problems.

Regarding the cooking test, we observed (Table 2) that 25% of the samples presented non-characteristic odor, described as rancid and unpleasant. It should be noted that all samples collected in Supermarket C showed a characteristic odor of cooked meat when evaluated in the cooking test; also, the yeast and mold counts of these samples were lower when compared to samples collected at other supermarkets. Similar results were found by Fernandes et al.<sup>30</sup> in the quality of the ground beef marketed in the city of Recife, when they verified that approximately 30% of the samples ( $n = 32$ ) presented altered sensory characteristics (color and odor).

Acero<sup>31</sup> reported that improperly stored meat favors the action of microorganisms and the development of acid, sulfide, and, finally, putrid odors. This phenomenon causes rejection on the part of consumers, since it causes the appearance of typically rancid flavors and odors, responsible for off flavors and off odors<sup>32</sup>.

At the beginning of the meat degradation process, the first gases released are ammonia and hydrogen sulfide<sup>33</sup>. Considering the results obtained in this study (Table 2), we observed that, for Nessler's test, 33.3% of the samples presented positive results, indicating the presence of ammonia, due to the breakdown of



Table 2. Qualitative parameters evaluated in ground beef.

Parameters	Supermarkets				Total number of samples (n = 12)
	A	B	C	D	
Cooking					
Positive	1 (8.3)	1 (8.3)	0 (0.0)	1 (8.3)	3 (25.0)
Negative	2 (16.7)	2 (16.7)	3 (25.0)	2 (16.7)	9 (75.0)
Nessler					
Positive	1 (8.3)	1 (8.3)	1 (8.3)	1 (8.3)	4 (33.3)
Negative	2 (16.7)	2 (16.7)	2 (16.7)	2 (16.7)	8 (66.7)
Eber (H <sub>2</sub> S)					
Positive	2 (16.7)	2 (16.7)	2 (16.7)	3 (25.0)	9 (75.0)
Negative	1 (8.3)	1 (8.3)	1 (8.3)	0 (0.0)	3 (25.0)
Sodium nitrate					
Positive	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Negative	3 (25.0)	3 (25.0)	3 (25.0)	3 (25.0)	12 (100.0)
Sodium nitrite					
Positive	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Negative	3 (25.0)	3 (25.0)	3 (25.0)	3 (25.0)	12 (100.0)
Sodium sulfite					
Positive	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Negative	3 (25.0)	3 (25.0)	3 (25.0)	3 (25.0)	12 (100.0)

\*H<sub>2</sub>S = hydrogen sulfide gas.

The results are expressed in n (%), where n is the number of samples we analyzed.

the proteins by microorganisms. Different results were obtained by Machi et al.<sup>16</sup>, when assessing the microbiological and physico-chemical parameters of ground beef. These authors verified that 100% of the analyzed samples (n = 30) were positive for Nessler's test, indicating that the beef under study was already undergoing proteolysis.

According to Silva Jr.<sup>34</sup>, ammonia gas may come from meat that has been stored for a long period under refrigeration, since psychotrophic and psychophilic microorganisms are mainly responsible for the production of this gas.

With regard to the H<sub>2</sub>S gas, we could verify that 75% of the samples were positive, indicating the presence of this gas. This is mainly due to the action of mesophilic microorganisms, usually in meats stored for a long time, since the sulfur amino acids of the meat are broken down, releasing sulfur, which will be used in the production of H<sub>2</sub>S gas<sup>35</sup>. Highlights to the samples collected in Supermarket D, as they were all positive for H<sub>2</sub>S gas, indicating that the sample was in a more advanced decomposition stage<sup>36</sup>.

Conceição and Gonçalves<sup>37</sup>, in a study of the physico-chemical quality of ground beef, verified that all beef samples (n = 20) were positive for the H<sub>2</sub>S gas test. However, Mesquita et al.<sup>35</sup> evaluated the physico-chemical quality of in natura beef approved at the reception of an industrial restaurant and verified that all the analyzed samples presented negative results for the H<sub>2</sub>S gas.

The H<sub>2</sub>S gas is mainly produced by mesophilic microorganisms, probably by prolonged exposure at room temperature. Moreover,

beef stored under poor conditions develops unpleasant odors, which are generated due to microbial action, because as the microbial population increases, so does the degree of proteolysis and, therefore, the production of H<sub>2</sub>S gas<sup>34,38</sup>.

Concerning fraud by the addition of sodium nitrite, nitrite and sulphite, 100% of the responses were negative, according to the legislation, which prohibits additives in fresh meat. When added, these preservatives can conceal the actual situation of the product, giving it a fresh appearance and suppressing occasional odors<sup>39</sup>.

Silva et al.<sup>40</sup>, when studying the presence of preservative additives in fresh meat, detected nitrite in samples of ground beef, with the highest concentration of nitrite found to be 1.17 and the lowest, 0.173 mg.Kg<sup>-1</sup>. Bonfada et al.<sup>41</sup> studied the presence of sodium sulfite and its influence on the physico-chemical and microbiological characteristics of cooled ground beef. They observed that of the 55 samples of cooled ground beef, two (3.63%) contained the sodium sulfite additive.

Preservatives are used to improve the sensory characteristics of the product, increase shelf life, inhibit the growth of pathogenic microorganisms and slow down oxidation<sup>42,43</sup>. However, the legislation prohibits the use of these additives in fresh meat<sup>5</sup>. Moreover, excess consumption of these additives has caused concern in the scientific community due to their harmful effects on human health<sup>44</sup>, related to the formation of carcinogenic chemical compounds, such as nitrosamines and nitrosamides<sup>45</sup>.



## CONCLUSIONS

Based on the results we obtained, we could verify that the ground beef sold in Erechim is not fully compliant with the required physico-chemical and microbiological parameters. This indicates the need to take immediate action in

partnership with the local health surveillance agency to improve the health education and the awareness of business owners and employees who work in meat packers and in food trade. With that, we can see to it that the products they market meet safety and quality requirements that protect the health of consumers.

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#### Conflict of Interest

Authors have no potential conflict of interest to declare, related to this study's political or financial peers and institutions.



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