

# Evaluation of the microbiological and physical-chemical quality of salted bovine meat marketed in establishments and free fairs in the north zone of Rio de Janeiro

## Avaliação da qualidade microbiológica e físico-química da carne bovina salgada comercializada em estabelecimentos e feiras livres na zona norte do município do Rio de Janeiro

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### ABSTRACT

**Introduction:** Salted beef is a highly handled product that can be contaminated by pathogenic microorganisms and foreign materials. **Objective:** Evaluate the microbiological and physical-chemical quality of *charque* and jerked beef. **Method:** Thirty samples were evaluated by microbiological research and count, physical-chemical analysis and microscopical research. **Results:** Halophilic bacteria count ranged from 5.2 to 8.8 log CFU/g, molds and yeasts from 2.8 to 8.2 log CFU/g. Although high results have been observed, there are not tolerance limits for these microorganisms in legislation. *Staphylococcus* spp. count ranged from < 2.0 to 7.8 log CFU/g, coliforms at 45 °C from < 3.0 to 9.3 x 10<sup>2</sup> MPN/g, beyond the absence of *Salmonella* spp. These results showed 11 samples above the limit for *Staphylococcus* coagulase positive and all samples were lower than the recommended for coliforms at 45°C. In the physical-chemical analysis, seven *charque* samples were observed above the limit recommended for the residual moisture content. In the analysis of fixed mineral residue, 18 *charque* samples and one of jerked beef were also above the limit. In the microscopical research, mites and furs were observed in three *charque* samples. **Conclusions:** Results showed the necessity of more effective hygienic-sanitary practices in the manipulation of these products.

**KEYWORDS:** *Charque*; Jerked Beef; Sanitary Surveillance; Quality Control

### RESUMO

**Introdução:** A carne bovina salgada é um produto altamente manipulado que pode ser contaminado por microrganismos patogênicos e materiais estranhos. **Objetivo:** Avaliar a qualidade microbiológica e físico-química do charque e *jerked beef*. **Método:** Trinta amostras foram analisadas por pesquisa e contagem microbiológica, análise físico-química e pesquisa microscópica. **Resultados:** Contagem de bactérias halofílicas variou entre 5,2 e 8,8 log UFC/g, bolores e leveduras entre 2,8 e 8,2 log UFC/g. Embora resultados elevados tenham sido observados, não há limites para esses microrganismos na legislação em vigor. A contagem de *Staphylococcus* spp. variou entre < 2,0 e 7,8 log UFC/g, coliformes a 45 °C entre < 3,0 e 9,3 x 10<sup>2</sup> NMP/g, além da ausência de *Salmonella* spp. Esses resultados mostraram 11 amostras acima do limite para *Staphylococcus* coagulase positivo e todas as amostras apresentaram-se inferiores ao recomendado para coliformes. Na análise físico-química, sete amostras de charque foram observadas acima do limite recomendado para o teor de umidade residual. Na análise de resíduo mineral fixo, 18 amostras de charque e uma de *jerked beef* também se apresentaram acima do limite. Na pesquisa microscópica, ácaros e pêlos foram observados em três amostras de charque. **Conclusões:** Os resultados mostraram a necessidade de práticas higiênicas-sanitárias mais eficazes na manipulação desses produtos.

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## INTRODUCTION

Food diseases are caused by the ingestion of food or water contaminated by pathogens, toxins produced by pathogens, chemicals, harmful objects or containing naturally toxic structures. Foods contaminated with biological agents and/or their toxins are the major cause of disease occurrence<sup>1,2</sup>. Normally, foods involved in outbreaks do not have sensory changes, being normal in appearance, odor and taste, which makes it difficult for the consumer to perceive the source of danger to health. Foods that have sensory alterations can hardly cause foodborne diseases because they are easily rejected by consumers<sup>3,4</sup>.

There are no official data about the consumption of these products in Brazil, but it is estimated that they are widely consumed. Most of the population identifies all variations of salted beef only as *carne seca*, although they differentiate not only in their technological processing, but also in their chemical composition and shelf life<sup>5,6</sup>. *Charque* is defined as meat product obtained from beef, with addition of salt and submitted to desiccation process. In turn, jerked beef is defined as meat product obtained from beef, added with salt and curing agents, and also submitted to desiccation process<sup>7</sup>.

Food analysis is an important tool, since it allows the characterization of physical-chemical and microbiological parameters of meat, thus favoring its quality control. The physical-chemical analysis is of great importance for the characterization, verification of compliance with the criteria established by the legislation and control of possible frauds. The technical and legal aspects of food require the standardization of parameters so that the analysis can be made assuring that the products are always in compliance with quality and identity standards<sup>8</sup>. The microbiological analysis of a food makes it possible to guarantee the safety and stability of the product through a qualitative and/or quantitative evaluation of the microbiota present, ensuring that the food matrix complies with the standards established by the legislation<sup>9,10</sup>. Foods, products directly related to health, as foreseen in the legislation<sup>11</sup>, have their control as competence of sanitary surveillance and, for this reason, it must use its tools to ensure that all products available to the population are in accordance with the standards required by the legislation, making them safe for consumption. The microscopical research is important because it allows checking the presence of possible foreign agents, including parasites<sup>12</sup>.

The objective of this study was to evaluate the microbiological and physical-chemical quality of salted bovine meat products marketed in neighborhoods of the northern Rio de Janeiro, to inform the community and health authorities about the hygienic conditions of meat products aforementioned.

## METHOD

Samples of salted meat products were purchased in small and large commercial establishments, as well as in free fairs, in the neighborhoods of the north zone in the city of Rio de Janeiro,

during March and April of 2017. A total of 30 indicative samples were collected, 27 from markets and three from free fairs, as shown in Figure, being 23 samples of *charque* and seven samples of jerked beef.

### Physical-chemical analysis

The analyses were done in triplicate of residual moisture content according to the gravimetric method of the oven at 105°C and fixed mineral residue by incineration in muffle at 550°C until the constant weight, both recommended by normative instruction (NI) n. 20, of July 21, 1999 from MAPA<sup>13</sup>.

### Microbiological analysis

The samples were submitted to the *Staphylococcus* coagulase positive count, enumeration of coliforms at 45°C and *Salmonella* spp. research, as recommended by RDC n. 12, of January 02, 2001 from Anvisa<sup>14</sup>. For a more complete analysis of the hygienic conditions, the counts of halophilic bacteria, molds and yeasts were also carried out. The methods used for the analysis were based on Compendium of Methods for the Microbiological Examination of Foods<sup>15</sup>. *Staphylococcus* coagulase positive count was based on the direct plate counting. Dilutions were done with 0.1% peptone saline and the culture media used was the Baird Parker agar. The coagulase test was performed using rabbit plasma. For enumeration of coliforms at 45°C, the adopted method was the most probable number (MPN). Dilutions were done with 0.1% peptone saline and the broth used was Rapid Hicoliform. The method adopted for *Salmonella* spp. research was based on the use of peptone saline solution as a pre-enrichment media, Mossel and Rappaport Vassiliadis broths as selective enrichment media and the Hektoen Enteric agar, Xylose Lysine Deoxycholate agar and *Salmonella* Differential agar as differential plating media. Confirmation was performed using biochemical and serological methods.

The count of halophilic bacteria, molds and yeasts was based on the direct plate counting. Dilutions were done with 3.0% peptone saline for halophilics and 0.1% peptone saline for molds and yeasts. The culture medium used for halophilic bacteria was



Figure. Distribution of sample collection points.



the Tryptone Soy agar with 3.0% sodium chloride (NaCl) and the Potato Dextrose agar for molds and yeasts.

### Microscopical research

Samples containing 50 g were weighed and placed in a beaker, immersed in 1,000 ml of distilled water for 30 min and homogenized every five min with the help of a glass stick. All the liquid was vacuum filtered using a Millipore filtration kit, a vacuum pump and GE Healthcare medium porosity filter papers (double ring quantitative filter paper, lot number FC005963). The filter papers were examined in Olympus SZ40 stereoscope for research of foreign particles<sup>16</sup>. Aiming to complement the method, researching possible microscopic parasites, the filter papers were immersed in 200 ml of distilled water after evaluation in the stereoscope, remaining in rest for 30 min. All the filtered material contained in the papers was removed and the solution was transferred to chalices for sedimentation. The volume was completed with distilled water when necessary and, after 30 min, with the help of a disposable Pasteur pipette, the pellet was collected and transferred to a glass slide and analyzed under an optical microscope. All the slides were completely examined<sup>17</sup>.

## RESULTS AND DISCUSSION

### Physical-chemical analysis

In the results obtained in the analysis of the determination of residual moisture content, none of the seven samples of jerked beef analyzed presented an average value higher than that prevailing in the current legislation (55%)<sup>18</sup>. Among the 23 samples of analyzed *charque*, seven presented a higher value than the allowed rates (45%)<sup>19</sup>. This represented 30.43% of the *charque* samples and 23.33% of the total samples. All the results found varied between 36.77% and 58.80% of residual moisture content.

According to Souza, Mano and Pardi<sup>20</sup>, an average of 52.6% was obtained for the residual moisture content among the 30 samples of *charque* analyzed, being 80% above the value specified in the legislation in force. Such result is above the one reached in this work. Likewise, Mársico et al.<sup>21</sup> analyzed 24 samples of *charque* and observed that 100% of the samples had a value higher than that established by the legislation.

Nishimoto et al.<sup>22</sup>, when analyzing 60 samples of jerked beef, found four samples with moisture levels higher than the expected, a result different from that found in the research. Similarly, Mascarenhas Filho and Silva<sup>23</sup> evaluated ten samples of jerked beef and found one sample above the expected rates. Correia and Biscontin<sup>24</sup> analyzed ten samples of *charque* and jerked beef and obtained values within the limits established by the legislation, which is also different from what was observed in this work. Santos and Hentges<sup>25</sup> analyzed three samples of *charque* and also observed that all presented the moisture content within the established limit.

The findings in this research can be considered unsatisfactory, since the determination of moisture content in salted meat products is of great importance because it is an intrinsic characteristic of food directly related to the proliferation capacity of microorganisms, including those capable of leading to disease development. Another important point is the fact that the moisture content above the allowed set up can be considered as fraud.

In the analysis of fixed mineral residue, a maximum value of 23.31% and a minimum of 13.06% were found. Of the seven analyzed samples of jerked beef, only one had a content of 19.09%, value above that established in current legislation (18.3%)<sup>18</sup>, which represents 14.29% of the samples of jerked beef analyzed and 3.33% of the total of samples. Among the 23 samples of *charque* analyzed, 18 were higher than established (15%). This represents 78.26% of the samples of *charque* analyzed and 60% of the total samples.

Mascarenhas Filho and Silva<sup>23</sup> in analyzing ten samples of jerked beef observed three values higher than those established by the legislation. In the same way, Santos and Hentges<sup>25</sup> analyzed three samples of *charque* and observed that all had content above the established limit. The results obtained in the present study corroborate with that observed by the authors. In contrast to the results found in this study, Correia and Biscontin<sup>24</sup> analyzed ten samples of *charque* and ten of jerked beef, and found a mean percentage of 17.25 ( $\pm 0.86$ ) and 18.07 ( $\pm 1.37$ ), respectively, all of which are in compliance with the legislation.

The high values found in this study can be explained by an excess of NaCl, as observed in some samples during the analysis, and also by the excess of curing salts. As described in the legislation, the use of curing salts in *charque* is prohibited, and more specific analyses are required to verify the presence of this substance in these products<sup>7</sup>.

### Microbiological analysis

The results obtained in the counts of halophilic bacteria were high and ranged from 5.2 log CFU/g to 8.8 log CFU/g. There was bacterial growth in all samples, observing colonies of mucoid aspect, varying between the white and yellow coloration. Abrantes et al.<sup>10</sup> analyzed 25 samples of *charque* and observed that the results ranged from 1.47 log CFU/g to 5.24 log CFU/g, which is lower than that observed in the present study.

Although the tolerance limit for halophilic bacteria is not determined in legislation<sup>14</sup>, the analysis has relevance since meat products submitted to the salting process have high concentrations of NaCl, making the medium conducive to the development of this group of microorganisms, which when in high number may favor the appearance of undesirable changes in the product<sup>26</sup>. Based on the results, which show high values found in the present study, they can be worrisome to maintain the quality during shelf life.

The analysis of fungi is also not mandatory in legislation and, consequently, no tolerance limits are established for these



microorganisms<sup>14</sup>. In all analyzed samples, significant growth of these microorganisms was observed, with values ranging from 2.8 log CFU/g to 8.2 log CFU/g. Costa and Silva<sup>27</sup> analyzed 96 samples of *carne-de-sol* and found values of  $3.8 \pm 1.02$  log CFU/g and  $4.44 \pm 0.95$  log CFU/g, lower than that found in the present study.

Based on the context of Jay<sup>28</sup>, the option to perform this analysis was due to the fact that these microorganisms are highly resistant and can survive and develop in inhospitable environments with high NaCl content and are frequently associated with deterioration of food. Another important factor, as reported by Jay<sup>28</sup>, is the ability of fungi to produce secondary metabolites detrimental to the health of humans and animals, the mycotoxins.

Of the 30 samples analyzed for *Staphylococcus* coagulase positive count, in only two (7.00%) no growth of *Staphylococcus* spp. was observed. Eighteen (60.00%) samples were coagulase positive and ten (33.00%) coagulase negative. Of the 18 samples tested positive for the coagulase test, 11 were above the established limit in the current legislation ( $5 \times 10^3$  CFU/g), representing a total of 66.70% of the coagulase positive samples and 40.00% of the total of analyzed samples. Of the 11 samples above the limit established for coagulase positive, eight were *charque* and three jerked beef.

Araujo et al.<sup>29</sup>, in a study in which seven samples of *charque* were analyzed, observed that all the samples were negative in the coagulase test and none were above the established limit for coagulase positive microorganisms. Rossi, Bonsaglia and Rall<sup>30</sup>, in the analysis of salty meats, observed that none of the ten samples analyzed were out of the established. The present study was not in agreement with the authors' findings. Nishimoto et al.<sup>22</sup> analyzed 60 samples of jerked beef and observed that in 58 samples the results were below the limit of detection of the technique used, one sample contained a count equal to  $1.2 \times 10^3$  CFU/g and another  $8.0 \times 10^3$  CFU/g. This last result was the only one found above the established limit, and was lower than the one observed in this work. Similarly, Abrantes et al.<sup>10</sup> analyzed 25 samples of *charque* and verified the presence of *Staphylococcus* spp. positive coagulase above the limits established by the legislation in five samples, ranging from 1.38 to 3.93 log CFU/g. In a study by Gurgel et al.<sup>31</sup>, 80 samples of meat were analyzed for *S. aureus*. The authors reported that only 21.25% of the samples were within the established standard, a result similar to that observed in the present study.

The fact that the product is highly manipulated from the acquisition and transportation, technological processing and sale, where they are fractionated and repackaged until reaching the consumer, explains the approximately three times above the limit samples of *charque*. It is known that these microorganisms are part of the normal microbiota of the human body, making the manipulator the main source of contamination<sup>28</sup>. The jerked beef is handled until the packaging stage, being handled again only by the consumer at the time of its preparation. This characteristic may be the justification for the reduced number of samples with

results above the recommended standard, which also ratifies the need for greater care with hygienic-sanitary practices during industrial processing, giving to the final product the necessary safety for consumption by the population.

Although there is no tolerance limit described for *Staphylococcus* spp. coagulase negative, the result is considered important since it is observed in the literature that there are reports of coagulase negative strains with the capacity to synthesize toxins in foods<sup>32</sup>.

In the enumeration analysis of coliforms at 45°C, the results of the samples were lower than those recommended by current legislation ( $10^3$  MPN/g)<sup>14</sup>. Rossi, Bonsaglia and Rall<sup>30</sup>, in a study aimed at characterizing the microbiological quality of salted meats, did not find results above the limits described and Abrantes et al.<sup>10</sup>, when analyzing 25 samples of *charque*, also did not find samples with MPN above the limit; similar results were found in this study. Different from what was observed in this study, Sousa<sup>33</sup> analyzed the microbiological characteristics of *carne-de-sol* and found samples with quantities of coliforms that are more than allowed in the legislation. In turn, Costa and Silva<sup>27</sup> analyzed the same product and observed the contamination by thermotolerant coliforms. Araújo et al.<sup>29</sup> analyzed seven samples of *charque* and verified that two presented contamination by thermotolerant coliforms. Similarly, Gurgel et al.<sup>31</sup> analyzed 80 samples of *carne-de-sol* and verified that 63.75% of the total samples analyzed were outside the established standard.

The findings are relevant and satisfactory for the quality of the evaluated samples, since the presence of bacteria of this group in foods indicates the possibility of contamination of fecal origin, besides the presence of pathogenic Enterobacteria, such as the *E. coli* pathotypes<sup>28</sup>.

In the results obtained in the *Salmonella* spp. research, the presence of the microorganism was not observed. These results corroborate with Santos and Hentges<sup>25</sup>, who analyzed three samples of *charque* and also observed absence of *Salmonella* spp. Similarly, Rossi, Bonsaglia and Rall<sup>30</sup> analyzed salted meats and did not observe the presence of these microorganisms. On the other hand, the study by Abrantes et al.<sup>10</sup>, in which 25 samples of *charque* were analyzed, showed the presence of *Salmonella* spp. in seven samples. Gurgel et al.<sup>31</sup>, when evaluating the quality of *carne-de-sol*, analyzed 80 samples and observed the presence of the agent in 25% of the total analyzed, different from the one obtained in this work.

The result of this study can be justified by factors such as the bacteria being considered a fastidious microorganism, not growing in environments in which water activity is less than 0.94. Regarding the saline concentration, these microorganisms do not tolerate high salt concentrations, being 9% a limit value. However, Jay<sup>28</sup> reported that another important factor is the presence of nitrite, effective in inhibiting the development of bacteria, which explains the absence in the jerked beef samples analyzed.



### Microscopical research

Foreign materials were observed in three *charque* samples, representing 10.00% of the total analyzed. Mites in one sample and furs in two were found. Santos and Rodrigues<sup>16</sup> analyzed 150 samples of salted meats and found foreign materials in 43.2%. Eggs, larvae and pupae, adult insects and fragments of insects, mites and rodent furs were observed. The results of this work are in agreement with the one observed by the authors. Likewise, Werneck<sup>34</sup> analyzed 117 samples of salted meat products and found 44 samples with foreign materials, representing 37.6% of the total analyzed. The furs observed in the two samples may indicate direct contact with rodents or faeces due to the habit of these animals to lick, ingesting hairs that are released along with the faeces, that are important sources of risk of contamination and render these foods unfit for consumption. Likewise, as observed by Santos and Rodrigues<sup>16</sup>, the presence of mites and furs in food makes the sample unfit for consumption and is also a source of public health risk.

Although this research was not carried out in the routine of quality programs in industries and the analysis of sanitary surveillance, the analysis was emphasized in the present work since the salted meat products, during their technological processing, are exposed in the environment for drying, which can lead to contamination. Also, *charque* is commonly sold in bulk at the points of sale, again being exposed to contamination. All samples in which foreign material was observed are of *charque*, which may indicate contamination during commercialization and/or manipulation in the establishments of sale. On the other hand, jerked beef is marketed in the same way as it is expedited by the industry, giving protection to the product until the arrival to the consumer. This indicates the need of greater care with the hygiene of premises and manipulation of these products, to guarantee the security to feed. Likewise, such care should also be taken in the industries during the technological processing, not only for *charque*, but also jerked beef, to reduce the risks to public health.

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### CONCLUSIONS

With the results observed in the work it is concluded that despite the absence of *Salmonella* spp. and reduced amount of coliforms at 45°C in the analyzed samples, the hygienic-sanitary quality is unsatisfactory due to non-compliance with the microbiological quality standards specified in the current legislation. In addition, the high rates of halophilic bacteria and fungi are worrisome, since there are reports in the literature related to such microorganisms to quality losses and outbreaks of foodborne diseases. It is inferred that it is necessary to adopt good practices and more hygienic-sanitary measures during the processing and marketing of the products in question, in order to reduce the contamination, since high rates of halophilic bacteria, *Staphylococcus* spp., molds and yeasts have been observed; as well as the presence of mites and furs.

In addition, the high residual moisture values found can facilitate the growth of pathogenic microorganisms and food deteriorating. The high number of *charque* fixed mineral residue above the limit is worrying, since the excess of mineral elements may indicate a possible fraud by the addition of curing salts and/or a greater amount of NaCl.

The results of the present research showed that the jerked beef samples had better quality parameters when compared to the *charque* samples, mainly because the commercialization of *charque* was done without due care at the points of sale, both in the free fairs and in the market. In contrast, jerked beef is normally marketed in vacuum packaging as it is expedited by the industries. It can serve as subsidies for the improvement of the service of the sanitary authorities and their commitment of a better performance. It also shows the need of investments in studies to elucidate the importance of the microorganisms whose limits of tolerance are not recommended in the legislation despite reports associating them to outbreaks of foodborne diseases.



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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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