

## Microbial contamination in Brazilian beef

### *Contaminação Microbiana na Carne Bovina Brasileira*

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#### ABSTRACT

Ensuring the quality of food products such as beef is extremely important to prevent outbreaks of food contamination caused by the presence of pathogenic bacteria, guaranteeing the physiological well-being of human beings, reducing the number of cases of contamination, and thus avoiding it becoming a public health problem. This study aimed to gather information from articles on microbial contamination in beef in Brazil using the scientometric method. Articles were searched using the academic platforms Scopus, Pubmed, and Scielo for keywords ("Escherichia coli" OR "Staphylococcus aureus" OR Listeria OR Salmonella OR "Lactobacillus Fermentum" OR Enterobacteria OR mesophiles OR Stecs) AND beef OR "carne bovina". The survey selected 69 articles that met the inclusion criteria for the study. The South, Southeast, and Center-West regions had the most studies evaluating the microbiological quality of beef. For the Southeast, this may be associated with the greater number of people living in the region and higher education institutions, while for the Midwest, it may be due to the potential for commercialization. Correlating the predominance of these two regions, they also had the highest number of articles analyzed, specifically São Paulo and Mato Grosso. We conclude that for beef quality to be guaranteed, preventive measures must be taken to monitor and improve sanitary conditions, from slaughter to marketing, so that products become increasingly reliable and do not pose a risk to public health.

**KEYWORDS:** Food safety. Pathogenic bacteria. Refrigerators. Scientometrics.

#### RESUMO

A garantia de qualidade de produtos alimentícios como a carne bovina, é de extrema importância para evitar surtos de contaminação alimentar devido a presença de bactérias patogênicas, garantindo o bem-estar fisiológico do ser humano, diminuindo o número de casos de contaminação, e consequentemente, deixando de ser um problema de saúde pública. O presente estudo visou realizar um compilado de informações em artigos sobre contaminação microbiana em carnes bovinas no Brasil, utilizando o método cienciométrico. As plataformas acadêmicas Scopus, Pubmed e Scielo foram utilizadas para o levantamento de artigos utilizando palavras-chaves ("Escherichia coli" OR "Staphylococcus aureus" OR Listeria OR Salmonella OR "Lactobacillus Fermentum" OR Enterobacteria OR mesofilos OR Stecs) AND beef OR "carne bovina". No levantamento foram selecionados 69 artigos, aos quais atenderam aos critérios inclusão do estudo. As regiões Sul, Sudeste e Centro-Oeste foram as que mais realizaram estudos avaliando a qualidade microbiológica em carne bovina. Para a região sudeste, isso pode ser associado ao maior número de pessoas alocadas na região e instituições de ensino superior, e em relação a região centro-oeste, deve-se ao potencial de comercialização. Correlacionando a predominância dessas duas regiões, também foram relacionados a elas os maiores números de artigos analisados, em específico São Paulo e Mato Grosso. Conclui-se que para que haja a segurança da qualidade da carne bovina, devem ser adotadas medidas preventivas, de fiscalização e melhoria de condições sanitárias, desde o abate a comercialização, para que o produto se torne cada vez mais confiável, e não um risco a saúde pública.

**PALAVRAS-CHAVE:** Segurança dos alimentos. Bactérias patogênicas. Frigoríficos. Cienciometria.

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## INTRODUCTION

Brazil is one of the largest beef producers in the world (RODRIGUES & MARTA-COSTA 2021). According to IBGE, 32.4 million cattle were slaughtered across the country in 2019, and beef production is expected to grow by 16.2% between 2019/20 and 2029/30 (MALAFAIA & BISCOLA 2023). Brazil is the world's second-largest producer, consumer, and exporter of meat products (HAHN 2020, REZENDE-DE-SOUZA et al. 2021). The growing increase in the production and consumption of meat products and their derivatives has raised food insecurity exponentially, mostly associated with bacterial contamination (REZENDE-DE-SOUZA et al. 2021). Meat products have both intrinsic and extrinsic factors that favor microbial growth in beef (SILVA et al. 2020a), such as high-water activity, favorable pH for most microorganisms, and high nutrient content (ALCANTARA et al. 2012, SILVA et al. 2020a).

Bacterial contamination in beef can be assessed through the physical and chemical conditions of the product, pH, for example, is a key physical and chemical parameter, as its alteration indicates the presence of deteriorating microorganisms in beef. According to Brazilian legislation Decree No. 9.013 of March 29, 2017 (BRASIL 2017), the ideal pH for meat consumption is between 5.4 and 5.8, with pH 6.4 being the critical limit for meat consumption, indicating a high level of microbial deterioration. It is worth highlighting that pH is directly linked to the physical characteristics of the product, such as color, water retention capacity, juiciness, and tenderness, reducing its shelf life and consumer acceptance (CORADINI et al. 2019, SALIM et al. 2017, SALIM et al. 2018).

The difference in meat color is linked to the concentration of the pigment myoglobin, the protein responsible for storing oxygen in the muscle (HENRIOTT et al. 2020, SHIN et al. 2021, WANG et al. 2021). Different forms of myoglobin cause differences in the color of meat, such as oxymyoglobin, which is bound to oxygen and provides a bright color; deoxymyoglobin, which is bound to water and has no oxygen present, providing a purplish color, commonly seen in vacuum-packed products; and metmyoglobin, which due to iron oxidation provides a brownish color (HENRIOTT et al. 2020, WANG et al. 2021).

Inspecting products of animal origin is crucial to guaranteeing the health of consumers, and food contamination must be monitored at all stages of production (ALCANTARA et al. 2012, REZENDE-DE-SOUZA et al. 2021, SILVA et al. 2020a). To this end, national reference laws and decrees have been created for the industrial and sanitary inspection of products of animal origin, such as Decree No. 9,013 of 2017, established by Law No. 1,283 of 1950, and Law No. 7,889/1989 (BRASIL 2017), which regulates and supervises production. For the marketing of meat products, slaughtered bovine production travels from farms to slaughterhouses and meatpacking plants. Slaughterhouses or meatpacking plants refer to the place where a series of controlled procedures are carried out to produce meat products (GÓMEZ et al. 2021, NESPOLO et al. 2014, PALMA et al. 2016). However, contamination by bacteria in fresh beef is still a challenge for public health, given the growing increase in food contamination (CARDOSO & CARVALHO 2006, GÓMEZ et al. 2021, MONTEIRO et al. 2013, MOREIRA et al. 2018) related to factors such as water, soil, and feed quality, poor

hygiene of utensils used in food preparation (REZENDE-DE-SOUZA et al. 2021), contamination by human and other animal feces, and atmospheric contamination (ABRANTES et al. 2014, FORSYTHE 2020, RIBEIRO JÚNIOR et al. 2021).

In Brazil, specific legislation for beef contamination by *Listeria* spp. remains undefined compared to other pathogens such as *Salmonella* spp. and *Escherichia coli*. *Listeria monocytogenes* is more strictly regulated in ready-to-eat foods, given its risk to public health (CAMARGO et al. 2016, GÓMEZ et al. 2021, SOARES et al. 2021). The National Health Surveillance Agency (ANVISA) and the Ministry of Agriculture, Livestock, and Supply (MAPA) are responsible for regulating food safety, including beef. The main rules and regulations include Resolution RDC No. 12 of January 2, 2001 (BRASIL 2001), which establishes microbiological standards for food. In this resolution, *Listeria monocytogenes* is mentioned in ready-to-eat foods, where it must not be present in 25 grams of the product. Furthermore, Normative Instruction No. 60 of December 23, 2019 (MAPA) defines the microbiological criteria and standards for products of animal origin, including beef, focusing on *Salmonella* spp. and *Escherichia coli* as indicators of hygiene and food safety.

Bacteria of the genera *Salmonella* and *Listeria* are the most prevalent in meat products (BIER et al. 2017, CORADINI et al. 2019, PERESI et al. 2021, SILVA et al. 2020, SOARES et al. 2021) and induce changes in the physicochemical and sensory properties of food, changing the pH and color (ROSSATO et al. 2010, SALIM et al. 2017, SALIM et al. 2018). Eating meat contaminated by *Salmonella* causes abdominal pain, diarrhea, and muscle pain. These symptoms characterize the disease known as salmonellosis. Typhoid fever, caused by *Salmonella enterica* serotype *typhi*, leads to more severe effects (CAPALONGA et al. 2014, CARDOSO & CARVALHO 2006). Ingesting meat contaminated by *Listeria monocytogenes* causes a food-borne illness called listeriosis (GÓMEZ et al. 2021, SILVA et al. 2016). Listeriosis can cause infections of the central nervous system, abortion, gastroenteritis, and septicemia, among other infections, even leading in certain cases to death, especially in children (MOABELO et al. 2023, WHITMAN et al. 2020).

The bacteria *Escherichia*, *Citrobacter*, *Enterobacter*, and *Klebsiella* comprise the coliform group and are directly associated with contamination in beef. Coliforms are divided into two groups: total coliforms and thermotolerant coliforms. Total coliforms are used to assess sanitary conditions, while thermotolerant coliforms provide good indicators of fecal contamination, as these organisms colonize the intestinal tract of warm-blooded animals (FERREIRA et al. 2014, MARQUEZINI et al. 2022, RISTORI et al. 2017, SALIM et al. 2017).

Since bacterial contamination in meat products originates from different sources (FERREIRA & FRANCO 2021), understanding the sources of bacterial contamination in meat products is critical, enabling action plans to be designed to reduce the levels of bacterial contamination that become a risk to public health. Thus, the systematic literature review method provides an overview of bacterial contamination in beef (BIER et al. 2022, DAMER et al. 2014) understanding the sources of bacterial contamination in meat products is critical, enabling action plans to be designed to reduce the levels of bacterial contamination that become a risk to public health. Thus, the systematic literature review method provides an overview of bacterial contamination in beef as an

informational process about the species and their related sources. This process entails quantitative methods to generate information on the subject (DONTHU et al. 2021).

The main objective of this systematic review of the literature is to gather information to improve the understanding of bacterial contamination in Brazilian beef. To this end, we aimed to analyze the bacteria that most contaminate beef, the Brazilian regions/states covered by the most studies; to analyze the environments in which beef is most exposed to contamination; analyze the types of physical and chemical parameters that most influence contamination; and to analyze the levels of bacterial contamination that pose a risk to public health.

## MATERIAL AND METHODS

### Searching the databases

We conducted a systematic literature review to gather scientific literature from 2012 to 2023. The search was carried out in the Scopus, PubMed, and Scielo databases under the keywords “*Escherichia coli*” OR “*Staphylococcus aureus*” OR *Listeria* OR *Salmonella* OR “*Lactobacillus fermentum*” OR *Enterobacteria* OR mesophiles OR Stecs AND beef OR “beef”. The search began on February 13, 2023, and a further search was performed on February 19, 2024, to include publications from 2023.

### Selecting the papers

The inclusion criterion consisted of selecting articles about bacteria in beef analyzed in Brazil, excluding grey literature (theses, dissertations, and monographs).

### Data analysis

For purposes of gathering data, we created an electronic spreadsheet containing the list of articles selected. The results were extracted by crossing lines and columns, with each column corresponding to the object-related answers, whereas the lines represented the articles. Each line referred to the articles selected and their respective data for individual analysis. The articles were analyzed from a macro to a micro perspective, starting from Brazil, advancing to cover the Brazilian regions and then states, to the environment where meat is exposed (slaughterhouses, meat-packing plants, and fairs), encompassing the bacteria examined, the list of bacteria against the physical and chemical parameters and the level of contamination in the meat in terms of the bacteria analyzed.

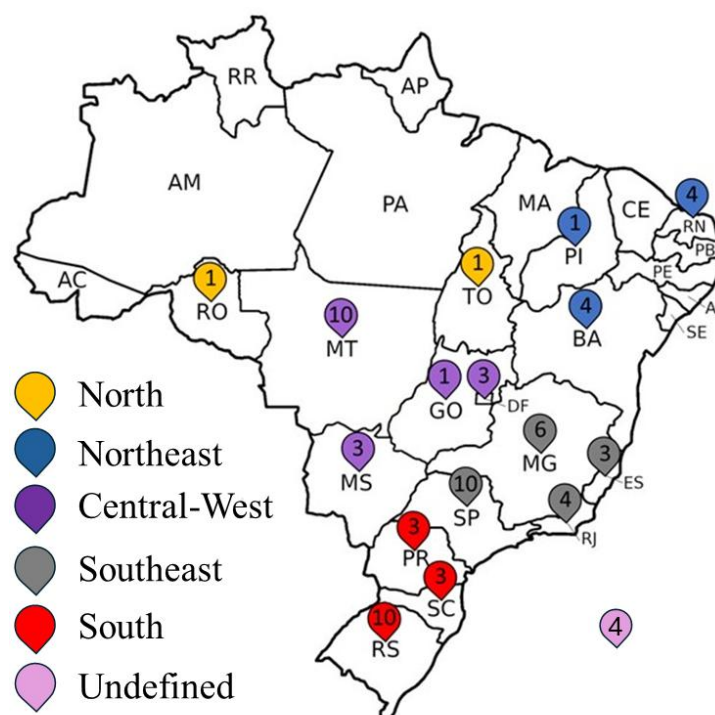
## RESULTS AND DISCUSSIONS

A total of 4,533 articles were retrieved, out of these, 3092 articles were selected once the exclusion criteria were applied. Following a careful analysis of the relationship with the theme, 69 articles were selected for this study.

### Analysis by region and state

The Southeast stood out among all Brazilian regions by the number of articles published addressing the presence of bacteria in beef (23 articles, 33.33%). Next, the Midwest (17 articles, 24.64%) and the South (14 articles, 20.29%) were also highlighted. Meanwhile, the Northeast (09 articles, 13.04%) and North (02 articles, 2.90%) (Figure 1) featured below ten articles published. However, four articles did not fit into any

particular region since gathered contamination surveys on different regions of the country.



**Figure 1.** Brazilian regions/states that have evaluated bacterial contamination in beef. The number within the markers shows the number of articles published in the state. The regions are organized by color, yellow: North Region; blue: Northeast; purple: Midwest; gray: Southeast; red: South; and pink: a combination of two or more regions.

### Analysis by beef exposure environments

Slaughterhouses and meat-packing plants accounted for the largest number of analyses of bacterial contamination in beef (33 articles), followed by supermarkets and butchers (28 articles). Meat display environments such as fairs, restaurants, bars, and public hospitals featured the least (Table 1).

**Table 1.** Exposure environments and analysis of beef contamination by bacteria. The number of articles and their frequency in percentages (%) were analyzed, considering the total of 69 published papers.

Environment	Number of articles	Frequency (%)
Slaughterhouses and meat-packing plants	33	47.82
Supermarkets and butchers	28	40.58
Fairs	04	5.80
Restaurants and Bars	03	4.35
Public Hospitals	01	1.45

Despite the intensive monitoring and caution concerning meat products, slaughterhouses, and meat-packing plants showed bacteria such as *Salmonella* spp., *Staphylococcus aureus*, total coliforms and thermotolerant, aerobic mesophilic, *Listeria* spp., acid lactic bacteria, and *Escherichia coli* – producer of the Shiga toxin (STEC).

### Bacteria analyzed

*Salmonella* spp. (23 articles; 22.77%) emerged as the most common contaminant in fresh beef, followed by *Listeria* spp (21 articles; 20.79%), *Escherichia coli* (14

articles; 13.86%), coliforms (10 articles; 9.90%), *Staphylococcus* (10 articles; 9.90%), aerobic mesophiles (09 articles; 8.91%), *Escherichia coli* O157: H7 and Shiga toxin-producing *Escherichia coli* (Stecs) (11 articles; 10.89%), *Klebsiella aerogenes* (02 articles; 1.98%), and lactic acid bacteria (01 article; 0.99%) (Figure 2).

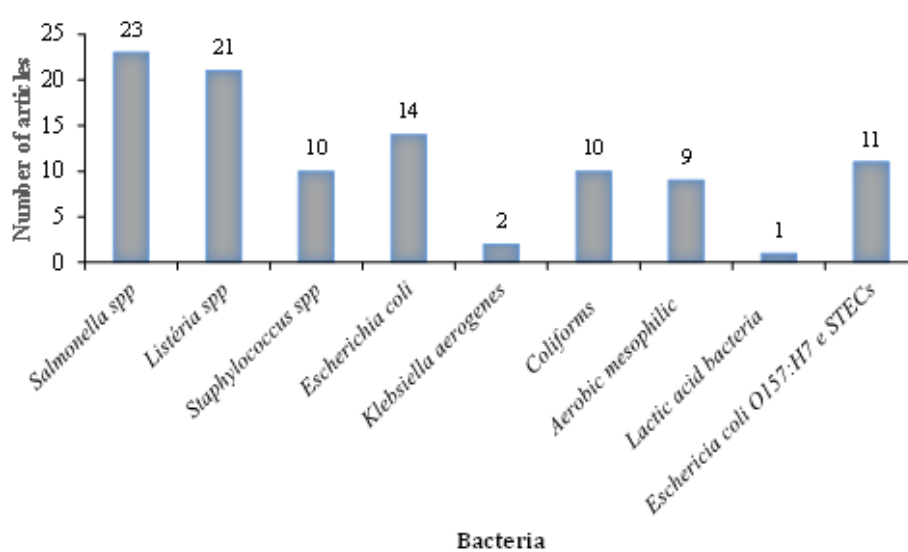


Figure 2. Relationship between the number of articles and the types of bacteria analyzed in the studies.

### Bacteria and their relationship with physical and chemical parameters

We analyzed the parameters of pH, color, temperature, water activity, and moisture. The parameters were analyzed as a set but not necessarily together, with pH being the main parameter analyzed (12.99% of the articles; Table 2).

**Table 2.** pH values analyzed in beef by environment in the papers analyzed. Analysis by paper considering the environments in which the meat was exposed and the pH of the meat analyzed in these environments. An asterisk (\*) indicates values below recommended and two asterisks (\*\*) represent values above recommended for human consumption according to Decree No. 9.013 of March 29, 2017.

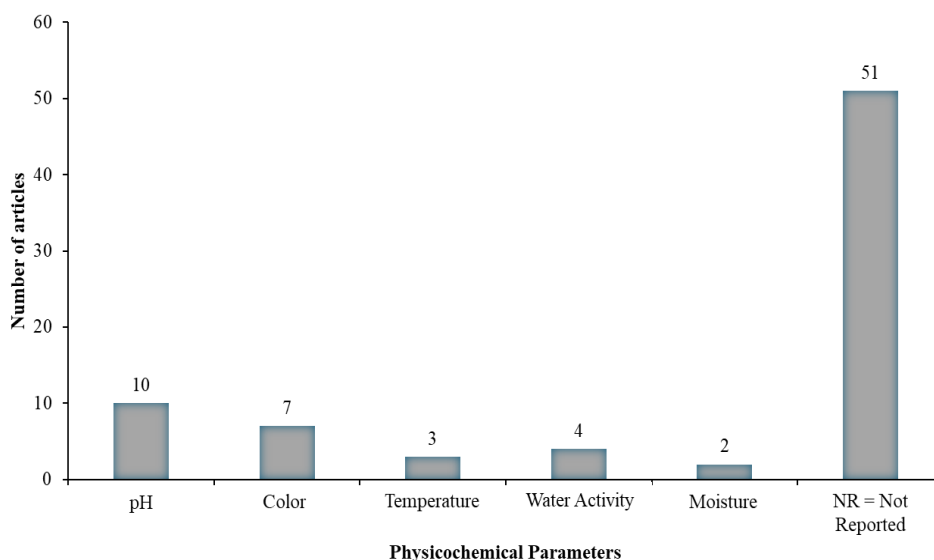
Papers	Author	Environment	pH analyzed
1	SILVESTRE et al. (2013)	Fairs and Supermarkets	5.55 – 5.84**
2	SAMPAIO et al. (2015)	Slaughterhouses	5.98** – 6.92**
3	VELHO et al. (2015)	Supermarkets	5.45 – 5.73
4	CAETANO et al. (2017)	Supermarkets	5.52 – 6.92**
5	CAMPELO et al. (2017)	Supermarkets	5.22* – 5.67
6	OLIVEIRA et al. (2017)	Butchers	5.11* – 6.95**
7	VENTURA et al (2020)	Butchers	5.72 – 5.89**
8	VIDAL JÚNIOR et al. (2020)	Butchers / Supermarkets	5.48 – 6.60** / 5.40 – 5.70
9	BARBOSA et al. (2022)	Restaurants	5.78
10	PEREZ et al. (2022)	Butchers	5.76 – 5.90**
Recommended pH – Decree No. 9.013, March 29, 2017			5.4 – 5.8

Only the studies by VELHO et al. (2015) and BARBOSA et al. (2022) showed pH values within the range recommended for human consumption by current legislation (Decree No. 9.013 of March 29, 2017). Meanwhile, the remaining studies showed pH values below or above those recommended for human consumption, making these



meats not recommended for consumption according to the legislation, since pH outside the recommended range indicates potential microbial proliferation.

Other physical and chemical properties assessed were color (07 articles, 9.09%), temperature (03 articles, 3.90%), water activity (04 articles, 5.19%), and moisture (02 articles, 2.60%). However, most articles (51 articles, 66.23%) did not include any analysis of physical and chemical parameters (Figure 3).



**Figure 3.** Analysis of physical and chemical parameters of Brazilian beef in the selected papers for the attributes pH, color, temperature, water activity, and moisture. Papers without this information are represented by NR = Not Reported. The values on the bars represent the number of papers analyzed.

Forty establishments were analyzed in one of the studies and only five of them had meat samples at a temperature suitable for commercialization according to the established standard of less than 7 °C. The highest temperature found in the studies corresponded to 25.5 °C (VENTURA et al. 2020).

### Contamination levels in brazilian beef

We could not determine the precise level of contamination, as most of the articles analyzed lacked information on the levels of bacterial contamination. Among the articles that did contain this information, few were expressed in terms of the amount of contamination per Colony-Forming Unit (CFU) and Most Probable Number (MPN) (Supplementary Material). Most articles only mention the presence or absence of the bacteria found, which may be associated with the microbiological analysis method used and the legislation in force in the year the study was published. Out of 27 articles that assessed contamination by *Salmonella* spp., 23 found positive results for this bacterium (BIER et al. 2022, BRASIL 2022). As for aerobic mesophilic bacteria, the articles that provided quantitative results mention some environments with values within the standard established by Brazilian legislation ( $10^6$  CFU.g<sup>-1</sup>) as well as those outside this standard. Only one article reported no bacteria present in the survey conducted.

All the papers that tested meat products for coliforms analyzed the presence of *Escherichia coli* bacteria and reported the presence and concentration of these

bacteria in the environment analyzed. Ten of these articles analyzed coliform counts, providing both quantitative (in CFU.g<sup>-1</sup>) and qualitative results (per MPN.100g<sup>-1</sup>) for thermotolerant coliforms and total coliforms.

## DISCUSSION

Although the Southeast has stood out, more recently the Midwest holds the greatest production and export of cattle in Brazil (IBGE 2023, TEIXEIRA et al. 2020), with production reaching around 75,175,767 animals (IBGE 2023). The export market has been increasingly demanding in terms of product quality. Although the export of Brazilian meat generates many resources for agribusiness, it can be hampered by the meat quality parameters required for export (AURÉLIO NETO 2018). Bacterial contamination can prevent Brazilian meat products from entering other countries. Each country has a checklist of quality standards for meat export. Currently, China is the major importer of Brazilian meat products, thus having agri-food relations with the Brazil-China Soy-Meat complex (ESCHER & WILKINSON 2019).

The Midwest region covers the states of Mato Grosso, Mato Grosso do Sul, Goiás, and the Federal District. All beef from the Midwest is exported via federal highways (BRs) until arriving at the port regions of Santos, in the state of São Paulo, and the port of Paranaguá, in the state of Paraná (ESCHER & WILKINSON 2019). Even though the state of Mato Grosso is the largest beef producer and exporter in the country, meat products still must travel through the export corridor in the Southeast and South. This may have added to the number of analyses of beef contamination in the Southeast and South regions.

The Southeast has the highest number of higher education institutions (MURAKAMI & FAUSTO 2013) and hence the greatest scientific production regarding the analysis of beef contamination. In turn, the Northeast and North have the lowest number of higher education institutions and the lowest production of beef, thus featuring bottom.

The state of São Paulo, in the Southeast, slaughtered more than 884,000 cattle in the second quarter of 2023. For this reason, it is ranked as the second largest beef exporter in the country, only behind the state of Mato Grosso, in the Center-West region (IBGE 2023). The southern state of Rio Grande do Sul has around 14 million cattle, placing it sixth in the country in terms of herd size (SILVA et al. 2014a, SOARES et al. 2021).

Slaughterhouses and meat-packing plants process meat *in natura*, in other words, the techniques involved should ensure that the product meets the requirements established by Brazilian legislation (BRASIL 2004, PALMA et al. 2016). These techniques consist of methods to prevent and delay changes in the natural conditions of meat, such as discoloration, decomposition, and putrefaction. These environments are focused on producing food that meets technical quality standards to comply with current legislation. *Escherichia coli* is part of the human and animal intestinal microbiota. However, pathotypes generically known as diarrheagenic *E. coli* (DEC), in high concentrations, cause intestinal infections (CASAGRANDE et al. 2013). Cattle are regarded as natural reservoirs of DEC, making them the main source of DEC contamination in food (NESPOLO et al. 2014). DEC can be transferred to the final



product when meat products are processed in slaughterhouses (CASAGRANDE et al. 2013, DAMER et al. 2014, FERIGOLO et al. 2021). Thus, these products are potential carriers of DEC to humans.

Supermarkets and butchers have a high risk of bacterial contamination due to the handling of products and utensils, the unsatisfactory hygiene practices of the handlers (ANDRADE et al. 2014, OLIVEIRA et al. 2017), as well as the storage conditions of meat products and the flow of people, all of which directly affect the contamination of beef (RIBEIRO JÚNIOR et al. 2021, SALIM et al. 2018, SILVA et al. 2020a, SILVA et al. 2020b). Fairs are also featured in the articles, in addition to supermarkets and butcher shops. Among all the environments analyzed (slaughterhouses and meat-packing plants, supermarkets, butchers, fairs, and restaurants), fairs pose the greatest risk of bacterial contamination (FILIOUSIS et al. 2009, VIDAL JÚNIOR et al. 2022). The flow of people and the poor facilities, especially in terms of basic sanitation and the protection of meat against contamination by pathogens circulating in the environment, lead to serious risks regarding food handling practices; there is also a lack of control over the proper storage temperature of the product; garbage is produced and accumulated nearby; all these factors together favor microbial contamination in this type of environment (EL-DEMERDASH & RASLAN 2019, FILIOUSIS et al. 2009).

The bacteria of the genera *Salmonella* and *Listeria* received the most attention, causing the diseases salmonellosis and listeriosis, respectively (CAPALONGA et al. 2014, CARDOSO & CARVALHO 2006, SILVA et al. 2016). These diseases are caused by food poisoning, which in severe cases can lead to death (CARDOSO & CARVALHO 2006, PERESI et al. 2021). For this reason, Brazilian legislation requires that *Salmonella* and *Listeria* be assessed (BRASIL 2001, SOUZA et al. 2014). The occurrence of these bacteria is associated with inadequate practices for obtaining, processing, and commercializing food. The most common forms of contamination are contact with the animal's skin, hair, gastrointestinal contents, the hands and clothes of employees, the water used to wash carcasses, and contaminated equipment and environments in slaughter and storage facilities (BIER et al. 2017, CAMARGO et al. 2019, DAMER et al. 2014). In addition, the presence of these bacteria induces changes in the physicochemical and visual properties of food, resulting in a change in pH and color (HENRIOTT et al. 2020, ROSSATO et al. 2010, SALIM et al. 2018, WANG et al. 2021). Despite the health risks associated with the presence of this bacterium (listeriosis: central nervous system infections, abortion, gastroenteritis, septicemia, and death), Brazilian legislation does not establish a microbiological standard for *Listeria monocytogenes* in fresh meat (BIER et al. 2022, BRASIL 2022).

The large-scale presence of the coliform group suggests unsatisfactory hygienic conditions that can be associated with any stage of the process, from obtaining the raw material to the final product, in inappropriate conditions of time and temperature throughout the production or preservation of the food (FERREIRA et al. 2014, PELAYO et al. 2019, SALIM et al. 2017). *Escherichia coli* is the main representative of the coliform group, which is also among the bacteria analyzed in the articles as a mandatory bacterium for evaluation according to Brazilian legislation (BRASIL 2022). *E. coli* can cause gastroenteritis, with symptoms such as abdominal pain, diarrhea, vomiting, nausea, or even blood in the stool (PELAYO et al. 2019).

According to the Normative Instruction No 12/2001 and the Brazilian legislation in force, the quantitative assessment of coliforms in meat products is mandatory (BRASIL 2022) for the presence of *E. coli* and other bacteria such as *Staphylococcus* spp, lactic acid bacteria, *Klebsiella aerogenes*, and *E. coli* producer of the Shiga toxin (STEC) (FERREIRA et al. 2014, RISTORI et al. 2017, TANABE et al. 2019).

Current legislation, Resolution No. 724 of July 1, 2022, fails to set standards for the analysis of thermotolerant coliform indicators (BRASIL 2022). However, given the recent change in legislation, the studies analyzed followed the recommendations of Resolution RDC No. 12 of January 2, 2001, for raw, chilled, or frozen meat products, which established a maximum limit of  $10^4$  NMP/g of sample, and by the National Commission of Norms and Standards for Food (1978), for raw meat, which are  $3.0 \times 10^2$  NMP/g for coliforms at 35 °C (Total) and  $5.0 \times 10^2$  NMP/g for coliforms at 45 °C (Thermotolerant), while for *E. coli* the standard set by the legislation in force is  $10^2$  CFU.g<sup>-1</sup> (BRASIL 2001). Analyses of coliforms, represented by *Escherichia coli*, are often used to assess microbiological quality, thus providing a good indication of compliance with Good Manufacturing Practices, Operational Health Programs, and the hygiene of equipment and utensils during operations (FERNANDES et al. 2017, SILVA et al. 2014b, VENTURA et al. 2020).

The count of aerobic or facultative mesophilic bacteria (35 - 37 °C) has been used as a microbiological indicator of food hygiene quality (CAMARGO et al. 2019). The authors found the cause to be cross-contamination when sanitizing the half carcasses. The evisceration process is among the most critical points of beef contamination, as the cut to remove the gastrointestinal tract can lead to incisions and leakage of intestinal contents, contaminating the carcasses (BIER et al. 2017, CASAGRANDE et al. 2013). The high concentration of total mesophilic aerobes in fresh meat reflects the microbiological quality of the marketed product, indicating deterioration and/or pathogenic microorganisms (DAMER et al. 2014, MACHADO et al. 2023, RIBEIRO JÚNIOR et al. 2021).

Bacterial contamination in beef can be assessed based on the physical and chemical conditions of the product. pH is directly related to physical properties such as color, water retention capacity, juiciness, and tenderness, reducing shelf life and consumer acceptance (CORADINI et al. 2019, SALIM et al. 2017, SALIM et al. 2018). Among the studies that measured pH, only two (Table 1, supermarket and restaurant) met the conditions established by Decree No. 9,013 of March 29, 2017 (pH = 5.4 to 5.8). The meats analyzed by SAMPAIO et al. (2015), CAETANO et al. (2017), and OLIVEIRA et al. (2017) exceed the critical limit (pH = 6.4) defined by the legislation (BRASIL 2017). This indicates that the meat sold from slaughterhouses (SAMPAIO et al. 2015) to supermarkets (CAETANO et al. 2017, OLIVEIRA et al. 2017) is putrefying. Although these meats are unfit for consumption, as they can cause serious gastroenteritis, they are still on the market. The change in pH, without considering the presence of bacteria, suggests two aspects: the proteolytic activity of endopeptidases in the meat itself or enzymes produced by microorganisms (ABRIL et al. 2023). Thus, pH cannot be an isolated parameter for assessing meat quality (OLIVEIRA et al. 2017, ROSSATO et al. 2010).

In addition to pH, color is a physical and chemical attribute that can be easily recognized visually. The changes that occurred in the color of the meat (the natural and ideal color of meat is bright red (SALIM et al. 2018)) were not associated with bacterial contamination, but with the use of acids and organic reagents to control microbial growth during the processing and storage of beef (HENRIOTT et al. 2020, SALIM et al. 2017, SALIM et al. 2018, WANG et al. 2021). This shows that commercialized meat is no longer a fresh product, but rather an altered item. Products of animal origin that should be fresh and are instead produced with adulteration violate all current legislation (BRASIL 2019, BRASIL 2017) and should not be offered for consumption.

Temperature is a key parameter in the preservation of meat, and the studies showed that most of the environments analyzed did not meet the standards established by law (BRASIL 2004, SALIM et al. 2017). This parameter is established by Ordinance No. 304/96 (BRASIL 1996), which establishes that beef, buffalo, and pork slaughterhouses must guarantee the distribution of these foods, standardized pieces, properly packaged and identified, at temperatures of up to 7 °C (BRASIL 1996). The authors report that high temperatures indicate a failure on the part of the establishments to monitor this parameter, which is essential for microbial growth; in addition, the temperature change was directly linked to long-term exposure to heat, without any type of refrigeration (BARBOSA et al. 2022, SAMPAIO et al. 2015, SILVESTRE et al. 2013, VELHO et al. 2015). This parameter was found to be altered in environments such as butchers and supermarkets, rising above the allowed level, suggesting that these establishments do not respect the standards established by legislation for marketing this product, thus favoring the proliferation of bacterial microorganisms and physical and chemical alterations.

Therefore, by comparing the articles analyzed, temperature appears as a factor for identifying poor-quality meat associated with the presence of *Salmonella* in most studies (Figure 3). It is Worth highlighting that the genera *Salmonella* grows at a wide range of temperatures, between 5°C and 45°C, with an optimum pH of around 6.5 to 7.5; thus, once not controlled, these factors favor the development of this bacteria (CAPALONGA et al. 2014, SILVA et al. 2020b).

Water activity and humidity are key parameters for this type of product since a high amount of free water favors microbial growth. A water activity higher than 0.83 allows for the growth of a large number of microorganisms, both spoilage and pathogenic, as well as affecting the visual appearance of the meat (SILVESTRE et al. 2013, VENTURA et al. 2020).

## CONCLUSION

This study gathered important information to enhance the understanding of bacterial contamination in Brazilian beef. Specifically, the following points are highlighted:

- 1) The Southeast stands out in the development of studies focused on controlling the contamination of beef marketed in Brazil. However, the Midwest, particularly the state of Mato Grosso, leads as the largest producer and exporter of beef worldwide, in an agri-food relationship of soy-beef-Brazil-China.

2) Slaughterhouses, meat-processing plants, supermarkets, and butcher shops are the most common sites for bacterial contamination of beef. The main bacteria associated with this contamination are *Salmonella* spp, *Listeria* spp, and *Escherichia coli*, and their presence is linked to failures in applying good hygiene practices, which compromises food safety. The recurrence of *Listeria* sp. and *E. coli* bacteria poses a public health risk. However, analyzing them as meat contaminants allows for a better understanding of their incidence and reinforces the need for preventive and control measures, considering that while *Listeria monocytogenes* is strictly controlled in ready-to-eat foods, its regulation in fresh beef is less stringent. This highlights the importance of good hygiene practices and control processes to ensure product safety.

3) Physical and chemical parameters should be analyzed in conjunction with other factors that directly influence both the proliferation and commercialization of beef, as they cause sensory changes, primarily associated with the appearance of the product. The lack of studies assessing the combination of the analysis environment, physical and chemical parameters, and bacterial concentration has limited the understanding of sources and forms of contamination in Brazilian beef. These findings underscore the need for further research and continuous improvement of control and prevention practices to ensure the quality and safety of beef in Brazil.

Supplementary Material - Contamination values for *E. coli*, *S. aureus*, *K. aerogenes*, Aerobic Mesophiles, and Lactic Bacteria, and Temperature and Environment, analyzed by the authors in the papers. [ ] 35 °C = concentration at 35 °C, [ ] 45 °C = concentration at 45 °C, NA = Not Analyzed, MPN.g<sup>-1</sup> = Most Probable Numbers per gram, CFU/cm<sup>2</sup> = Colony Forming Unit per square centimeter, CFU.g<sup>-1</sup> = Colony Forming Unit per gram.

Nº	Paper	Environment	<i>E. coli</i>	<i>S. aureus</i>	[ ] 35 °C	[ ] 45 °C	<i>K. aerogenes</i>	Aerobic mesophilic bacteria	Lactic acid bacteria
1	Bier et al. (2022)	Butchers, supermarkets	NA	14.8X10 <sup>3</sup> - 11X10 <sup>3</sup> 33X10 <sup>3</sup> - 2.0X10 <sup>4</sup> 30X10 <sup>3</sup> - 18X10 <sup>4</sup> 9.2X10 <sup>3</sup> - 43X10 <sup>3</sup> 50X10 <sup>3</sup> - 21X10 <sup>3</sup> 17X10 <sup>3</sup> - 21X10 <sup>3</sup> 43X10 <sup>3</sup> - 10X10 <sup>3</sup> 19X10 <sup>3</sup> - 21X10 <sup>3</sup> 91X10 <sup>3</sup> - 47X10 <sup>3</sup> CFU.g <sup>-1</sup>	NA	NA	NA	NA	NA
2	Abrantes et al. (2014)	Meat-packing plants	NA	NA	NA	0,36 NMP.g <sup>-1</sup>	NA	NA	NA
3	Silva et al. (2020)	Slaughterhouse	NA	NA	NA	NA	NA	4.33 and 5.71 log CFU.g <sup>-1</sup>	3.17 and 3.53 log CFU.g <sup>-1</sup>
4	Camargo et al. (2019)	Meat-packing plants	3.06, 1.58, 1.89, 1.53 log CFU/cm <sup>2</sup>	NA	3.20, 1.73, 2.05, 1.81 log CFU/cm <sup>2</sup>	NA	3.27, 1.79, 2.20, 2.11 log CFU/cm <sup>2</sup>	4.51, 2.47, 2.64, 2.93 log CFU/cm <sup>2</sup>	NA
5	Casagrande et al. (2013)	Meat-packing plants	1.25, 0.58, 0.29, 2.71, 0.46, 0.71, 1.57, 0.23, 3.17, 14.06, 3.26, 0.08 log CFU/cm <sup>2</sup>	NA	NA	NA	NA	NA	NA
6	Da Silva et al. (2014)	Meat-packing plants	0.31 to 5.07 log CFU/100 cm <sup>2</sup> 2.42 log CFU/100 cm <sup>2</sup> 2.10 log /100 cm <sup>2</sup>	NA	NA	NA	NA	NA	NA

7	De Souza Velho et al. (2015)	Supermarkets	NA	NA	3.75 – 1.82	2.36 – 0	NA	NA	NA
					3.22 – 3.61 – 3.70 – 5.25 – 5.18 – 3.96 – 3.69 – 3.7 – 4.07 – 3.77 – 5.32 – 4.65 – 5.92 5.75 MPN.g <sup>-1</sup>	1.55 – 1.65 2.1 – 5.15 3.6 – 2.54 0.87 – 3.3 1.91 – 1.87 2.33 – 1.61 5.25 – 4.84 MPN.g <sup>-1</sup>			
8	Da Silva Silvestre et al. (2013)	Fairs and supermarkets	NA	4.87 – 2.38 - 4 CFU.g <sup>-1</sup>	NA	4.08 – 3.31 – 3.74 NMP/g	NA	5.41 - 3.35 - 4.6 log CFU.g <sup>-1</sup>	NA
9	Oliveira et al. (2017)	Butchers	46.66% (28/60) of the samples	2.4 x 10 <sup>2</sup> to 8.2 x 10 <sup>5</sup> CFU.g <sup>-1</sup> <i>Staphylococcus</i> sp	28.33% (17/60) above 10 <sup>3</sup> MPN.g <sup>-1</sup>	4/60 (6.66%) above 10 <sup>3</sup> MPN.g <sup>-1</sup>	NA	2.0 x 10 <sup>3</sup> to 3.42 x 10 <sup>6</sup> CFU.g <sup>-1</sup>	NA
10	Vidal Junior et al. (2020)	Fairs and supermarkets	3.69 to 4.60 – 3.00 to 4.04 log CFU.g <sup>-1</sup>	NA	4.30 to 5.67 – 3.60 to 5.30 log CFU.g <sup>-1</sup>	NA	NA	NA	NA
11	Campelo et al. (2017)	Supermarkets	NA	NA	43 MPN.g <sup>-1</sup>	NA	NA	6.88 log CFU.g <sup>-1</sup>	NA
12	Fernandes et al. (2017)	Meat-packing plants	NA	NA	NA	1.7 – 2.4 – 3.0 – 2.90 – 2.90 - 1.7 – 2.1 MPN.g <sup>-1</sup>	NA	NA	NA

## NOTES

### AUTHOR CONTRIBUTIONS

Conceptualization, methodology, and formal analysis, Izabela Marques Sousa, Karina da Silva Chaves and Danielle Regina Gomes Ribeiro-Brasil; software and validation, Izabela Marques Sousa and Danielle Regina Gomes Ribeiro-Brasil; investigation, Izabela Marques Sousa, Karina da Silva Chaves and Danielle Regina Gomes Ribeiro-Brasil; resources and data curation, Izabela Marques Sousa, Karina da Silva Chaves and Danielle Regina Gomes Ribeiro-Brasil; writing-original draft preparation, Izabela Marques Sousa, Karina da Silva Chaves and Danielle Regina Gomes Ribeiro-Brasil; writing-review and editing, Izabela Marques Sousa, Karina da Silva Chaves and Danielle Regina Gomes Ribeiro-Brasil; visualization, Izabela Marques Sousa, Karina da Silva Chaves and Danielle Regina Gomes Ribeiro-Brasil; supervision, Karina da Silva Chaves and Danielle Regina Gomes Ribeiro-Brasil; project administration, Karina da Silva Chaves and Danielle Regina Gomes Ribeiro-Brasil; funding acquisition, not applied. All authors have read and agreed to the published version of the manuscript.

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Not applicable for studies not involving humans or animals.

### INFORMED CONSENT STATEMENT

Not applicable because this study did not involve humans.

### DATA AVAILABILITY STATEMENT

The data can be made available under request.

## CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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