




## Hygienic sanitary conditions of vacuum packed beef produced by slaughterhouses qualified for export in the Mato Grosso state, Brazil

Maxsueli Aparecida Moura Machado<sup>1</sup> Barbara Müller<sup>2</sup>  
Ricardo César Tavares Carvalho<sup>2</sup> Eduardo Eustáquio de Souza Figueiredo<sup>2,3\*</sup> 

<sup>1</sup>Graduação em Ciência e Tecnologia de Alimentos, Faculdade de Nutrição, Universidade Federal de Mato Grosso (UFMT), Cuiabá, MT, Brasil.

<sup>2</sup>Programa de Pós-graduação em Nutrição Alimentos e Metabolismo, Universidade Federal de Mato Grosso (UFMT), Cuiabá, MT, Brasil.

<sup>3</sup>Programa de Pós-graduação em Ciência Animal, Universidade Federal de Mato Grosso (UFMT), Av. Fernando Correa da Costa, 2367, 78060-900, Cuiabá, MT, Brasil. E-mail: [figueiredoeduardo@ufmt.br](mailto:figueiredoeduardo@ufmt.br). \*Corresponding author.

**ABSTRACT:** Brazil is the largest exporter of beef of the world and Mato Grosso State is the highest beef producer in this country. To maintain product competitiveness and market expansion, sanitary hygienic control of the entire process is indispensable to ensure the attainment of harmless beef and quality. The objective of this study was to evaluate the hygienic sanitary conditions of vacuum-packed beef produced by establishments qualified for export in the state of Mato Grosso, Brazil. A total of 60 samples were submitted to coliforms counts at 35°C and at 45°C and *E. coli*. The mean contamination by at 35°C and coliforms at 45°C were  $3,1 \times 10^2$  MPN/g and 7.7 MPN/g respectively. The presence of *E. coli* was verified in five samples, representing an occurrence of 8.3% (5/60), and *Salmonella* spp. in 5% (03/60) of the analyzed samples. The MPN (Most Probable Number) average of coliforms at 35°C and 45°C are in accordance to national and international legislation; however, the presence of *Salmonella* spp., *E. coli* in some sample indicates a low risk of occurrence of salmonellosis and colibacillosis transmitted by the evaluated beef. However, transmission risk of these diseases cannot be ruled out, since the presence of *E. coli* does not depend on the amount of coliforms and national legal standards established for the group of thermotolerant coliforms.

**Key words:** coliforms, *Escherichia coli*, *Salmonella* spp., meat.

## Condições higiênicas sanitárias de carne embalada a vácuo produzida por abatedouros frigoríficos habilitados a exportação no estado de Mato Grosso, Brasil

**RESUMO:** O Brasil é o maior exportador de carne bovina do mundo e Mato Grosso é o maior produtor de carne bovina do país. Para manter a competitividade do produto e a expansão do mercado, o controle higiênico sanitário de todo o processo é indispensável para garantir a obtenção de carne inócua e de qualidade. O objetivo desse estudo foi avaliar as condições higiênicas sanitárias da carne bovina resfriada embalada a vácuo, não maturada produzida por estabelecimentos detentores de Serviço de Inspeção Federal (SIF) no estado de Mato Grosso. Um total de 60 amostras foram submetidas a contagem de coliformes à 35°C, coliformes à 45°C, pesquisa de *E. coli* e *Salmonella* spp. A contaminação média por coliformes à 35°C e coliformes à 45°C da carne bovina mato-grossense inspecionada pelo SIF foi  $3,1 \times 10^2$  NMP/g e 7,7 NMP/g, respectivamente. A presença de *E. coli* foi verificada em cinco amostras o que representou uma ocorrência de 8,3% (5/60) e *Salmonella* spp. em 5% (3/60). Foi avaliado que a média NMP de coliformes à 35°C e à 45°C atende a legislação nacional e internacional, no entanto, a verificação da presença de *Salmonella* spp., *E. coli* em algumas amostras, evidenciou um baixo risco de ocorrência de salmonelose ou colibacilose transmitidas pela carne. Ainda assim, não se pode descartar o risco de veiculação dessas doenças, uma vez que a presença de *E. coli* independe da quantidade de coliformes e dos padrões legais nacionais estabelecidos para o grupo dos coliformes termotolerantes.

**Palavras-chave:** coliformes, *Escherichia coli*, *Salmonella* spp., carne bovina.

Brazil is the second largest producer of beef in the world, being surpassed only by the USA. Meanwhile, Brazil is the largest exporter of this product, with supply to more than 100 countries, the Mato Grosso State is the highest meat producer in Brazil (ABIEC, 2016; BRASIL, 2016). To maintain product competitiveness and market expansion, sanitary hygienic control of the entire

process is indispensable to ensure the attainment of harmless beef and adequate quality. This is currently especially true because of the recent events of the so-called “weak meat operation” that raised doubts about Brazilian meat safety. Thus, studies are being conducted to characterize and evaluate the contamination of this type of product in Brazil (SANTOS, et al. 2017).

Brazil's Resolution RDC N° 12 (BRASIL, 2001) establishes microbiological standards for food; however, it does not establish references for coliforms at 35°C and *E. coli* in vacuum-packed, fresh-frozen beef. Conversely, Brazilian beef importers demand the evaluation of these quality indicators.

Currently, some Brazilian slaughterhouses were authorized to sell fresh chilled and frozen beef to the USA. Therefore, it is extremely important to evaluate these products, as these buyers, as well as Brazil, demand the total safety of beef, since these foods are related to the presence of pathogenic microorganisms, such as *Salmonella* spp. and *Escherichia coli*, originated from fecal contamination. In this regard, there is need for further studies devoted to the study of the microbiological profile of packaged vacuum beef, since the detection of these microorganisms in indispensable product safety. Thus, in this study, the hygienic sanitary conditions of vacuum-packed cooled fresh beef, produced by slaughterhouses in the state of Mato Grosso qualified for export to different countries of the world were evaluated.

A convenience sample was established according to the number of bovine abattoirs under permanent supervision by the Brazilian Federal Inspection Service (SIF) in the state of Mato Grosso. Thus, a total of 60 samples of fresh beef packaged under vacuum, cooled and not mature, were obtained from nine cattle slaughtering establishments, all authorized for export. The samples were cooled to between 1°C and 7°C, had average weight of 1.5kg, and were composed of individual bovine meat cuts such as shoulder clod (*triceps brachii*), brisket (*pectoralis superficialis*), roast beef (*longissimus dorsi*), eye cap (*trapezius thoracis*), rump cap (*Biceps femoris*), rump (*gluteus medius*), eye of round (*semitendinosus*) and tenderloin (*psoas major*).

Samples were submitted to microbiological analyzes, in triplicate, for quantification of coliforms at 35°C, coliforms at 45°C, *E. coli* and *Salmonella* spp., based on the APHA method (KORNACKI & JOHNSON, 2001). Suspected *Salmonella* spp. colonies were also subjected to Polymerase Chain Reaction (PCR) to amplify the *hlyA* gene, that is genus-specific and one of the main transcriptional regulators of other invasion genes (GUO et al., 2000; BAJAJ, et al., 1995). Results were interpreted according to the indication of each method and compared with the microbiological references and standards established by Resolution RDC N°. 12 (BRASIL, 2001).

The average beef coliform contamination at 35°C and 45°C were  $3.1 \times 10^2$ MPN/g and 7.7MPN/g, respectively. The presence of *E. coli* was observed

in five samples, representing an occurrence of 8.3% (5/60) and *Salmonella* spp. in 5% (03/60) of the analyzed samples. The mean coliforms at 35°C and coliforms at 45°C were calculated according to the 60 analyzed samples, and only samples with coliforms counts in the MPN table or showing the presence of *E. coli* and *Salmonella* spp. are listed in table 1.

Considering these results, a low average count of coliforms at 45°C was observed, according to ANVISA's sub item h of RDC N°. 12. Low occurrence of *E. coli* was also detected; although, some samples (5875, 7627 and 7649) exceeded the maximum limit for coliforms at 45°C (BRASIL, 2001).

The average coliforms counts at 35°C and 45°C were lower than other studies that evaluated the microbiological quality of beef produced and marketed in different Brazilian states (OLIVEIRA et al., 2002; OLIVEIRA et al., 2008; ROSINA et al., 2013). This contamination is directly related to GMP, Standard Operating Procedures (SOP), and Hazard Analysis and Critical Control Points (HACCP), considering that coliform samples with MPN above  $10^3$  were from different slaughtering establishments, many of them were not eligible for export (Table 1). Thus, it is possible to verify the natural variation between application of existing quality program tools between the different slaughtering establishments.

*E. coli* was detected in some analyzed samples (3413, y011, 7627, 7649, 2569). *E. coli* beef research is required by several meat importing countries as a quality and food safety indicator. The occurrence of *E. coli* in vacuum-packed beef is independent of MPN/g of coliforms at 45°C, but its presence indicates fecal contamination, possible existence of other pathogens and the risk of colibacillosis transmission by pathogenic *E. coli* (GILL, 1998). This was also observed in slaughtering establishments with federal and provincial registration in Canada, where a significant statistical correlation between coliform counts and the presence of *E. coli* was not verified (JONES et al., 2014).

The presence of *Salmonella* spp. was verified in the samples 2573, 3587 and 2547 representing an occurrence of 5%, in disagreement with the Brazilian legislation (BRASIL, 2001) making those samples unfit for human consumption. Other studies carried in the state Mato Grosso on both meat from butchers and vacuum packed beef, detected a *Salmonella* spp. occurrence of 17% and 5.6% of respectively (SOUZA et al., 2012).

The presence of *E. coli* and *Salmonella* spp. identified in the samples of the present study came from four and two different slaughtering

Table 1 - Coliform counts, *E. coli* and *Salmonella* spp. research on cooled fresh beef, produced by slaughterhouses qualified for export in the Mato Grosso State, Brazil.

Sample**	Total Coliforms (MPN/g)	Coliforms 45°C (MPN/g)	<i>E. coli</i>	<i>Salmonella</i> spp.	Slaughterhouses
3586	<3,0	9,2	A	A	I
3412	3,6	<3,0	A	A	I
3587	3,6	<3,0	A	P*	I
3407	3,6	3,6	A	A	I
3585	3,6	3,6	A	A	I
2573	3,6	3,6	A	P*	I
3408	7,4	3,6	A	A	I
1343	15	3,6	A	A	I
2566	23	<3,0	A	A	I
3583	23	23	A	A	I
2571	43	43	A	A	I
2568	43	<3,0	A	A	I
7443	>1,1x10 <sup>3</sup>	<3,0	A	A	I
2572	4,6x10 <sup>2</sup>	9,2	A	A	I
3413	15	1,5	P	A	I
2569	93	23	P	A	I
2907	3,6	3,6	A	A	II
7649	>1,1x10 <sup>3</sup>	>1,1x10 <sup>3*</sup>	P	A	II
2902	3,6	<3,0	A	A	III
X002	3,6	<3,0	A	A	IV
y009	9,2	3,6	A	A	IV
y012	9,2	<3,0	A	A	IV
y010	23	23	A	A	IV
y013	23	3,6	A	A	IV
X004	44	<3,0	A	A	IV
X001	93	<3,0	A	A	IV
X003	1,1x10 <sup>3</sup>	3,6	A	A	IV
X006	1,1x10 <sup>3</sup>	<3,0	A	A	IV
y014	1,1x10 <sup>3</sup>	1,1x10 <sup>3*</sup>	A	A	IV
X005	>1,1x10 <sup>3</sup>	3,6	A	A	IV
y011	9,2	9,2	P	A	IV
5875	>1,1x10 <sup>3</sup>	>1,1x10 <sup>3*</sup>	A	A	V
7627	>1,1x10 <sup>3</sup>	>1,1x10 <sup>3*</sup>	P	A	VI
2547	<3,0	<3,0	A	P*	VII
X***	3,1x10 <sup>2</sup>	7,7			
S	4,7x10 <sup>2</sup>	9,3			

X = Average; S = standard deviation; A = absence; P = presence; MPN = Most probable number per gram; \*Exceeded limit in accordance to subitem h of RDC No. 12 (BRASIL, 2001); \*\*Only samples with counts above 3.0 MPN for coliforms, presence of *E. coli* or *Salmonella* spp.; \*\*\*Average calculated according to 60 samples.

establishments respectively (Table 1), and, especially regarding *Salmonella* spp., most underwent SIF inspection. In this regard, studies that identify and report these results are extremely necessary, since the presence of these pathogens makes products unacceptable and interferes with the purchase of these foods by other countries.

Thus, it was evaluated that MPN average of coliforms at 35°C and coliforms at 45°C are in

attendance to national and international legislations; although, the presence of *E. coli* in the groups of coliforms at 45°C is not related to the amount of coliforms or legal standards, which does not rule out the risk of transmission of this pathogen and its sorogroups. Likewise, the presence of *Salmonella* spp. in some samples indicated that; although low, the risk for dissemination of diseases such as salmonellosis exists.

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## DECLARATION OF CONFLICTING INTERESTS

None of the authors of this paper has a financial or personal relationship with other people or organizations that could inappropriately influence or bias the content of the paper.

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