Original Paper

Prevalence of some Foodborne Pathogens (FBPs) in imported chilled and frozen beef in Egypt

Aya M. A. Ahmed[1,2], Faten S. Hassanin[1], Gehan Sayed A. Afify[1], Shimaa.N. Edris[1*]

[1] Department of Food Hygiene and Control, Faculty of Veterinary Medicine, Benha University
[2] National Food Safety Authority, Cairo, Egypt

ARTICLE INFO

ABSTRACT

Food safety is a global priority due to foodborne pathogens contamination. This study determined the prevalence of Aerobic mesophilic bacteria count (AMB), *Staphylococcus aureus*, *Salmonella spp.*, *E. coli* and *Listeria monocytogenes* in imported chilled and frozen beef in Egypt. The experimental results revealed that the AMB (cfu/g) in chilled samples varied from 5.3×10^4 to 4.7×10^6, with an average of $8.35\times10^5\pm1.06$. In contrast, for frozen samples, the range was from 9.7×10^3 to 1.5×10^6, with an average of $3.11\times10^5\pm0.29$. *Staphylococcus aureus* was found in 24 % of chilled beef samples and 16 % of imported frozen beef samples. The prevalence of Salmonella spp. was identified in 5% of chilled beef samples and 3% of imported frozen beef samples . Additionally, *Escherichia coli* was identified in and 14% of chilled samples and 8% of frozen samples. The presence of *L. monocytogenes* was identified in 3% of chilled beef samples and 1% of frozen samples. The results obtained validated the inadequate bacteriological quality of certain imported chilled and frozen meats sold in the markets of Cairo and Qalyubia. This quality deficiency is a result of unclean transportation practices that continue to the retail levels. Chilled comprehensive findings, it can be inferred that imported chilled and frozen beef poses a substantial bacteriological public health risk and requires specific control measures.

1. INTRODUCTION

Nowdays, foodborne pathogens (FBPs) pose a substantial treat to public health. Meat is regarded as the most essential dietary item due to its nutritional value and palatability. Due to its pH, moisture, fat, protein, and fermentable carbohydrates, raw beef is ideal for spoiling and hazardous bacteria growth (Kebede and Getu, 2023). Foodborne illnesses are the primary cause of death and infectious diseases in developing countries. Bacteria cause 90% of foodborne infections, making them the biggest microbiological concern. By regulating potential contamination sources during harvesting, processing, distribution, retail markets, food service outlets, and the home, raw meat health concerns can be reduced. The microbiological quality of meat depends on proper slaughtering, sanitary processing, cold chain storage during and after processing, and hygienic retail handling (Borch and Arinder, 2002). The meat industry must constantly ensure product quality and safety. Improper storage, shipping, or handling can exacerbate microbial infection. Freezing and refrigeration are popular strategies used to inhibit the growth of microorganisms that cause food-borne illnesses (Mohammed et al., 2021). However, cooled and frozen imported beef poses serious health risks and requires careful control. Many studies have shown that chilled and frozen storage length is vital to meat quality and preventing deterioration during export and other activities (Leygonie et al., 2012). Egypt imports a variety of meats to meet animal protein demands. A common bacterium, salmonella, causes food poisoning. Raw meat is a major source of these bacteria, which can cause foodborne illnesses. Salmonellosis

causes stomach flu (gastroenteritis). This illness causes nausea, vomiting, stomach cramps, and bloody diarrhea. Fever, headache, and myalgia occur. Fluid loss can cause dehydration, especially in babies and the elderly (Ehuwa et al., 2021). *Escherichia coli* is the most reliable faecal infection surrogate (Xu et al., 2022). *E. coli* can cause a variety of enteric and extraintestinal illnesses in addition to its commensal role (Manges and Johnson, 2012). Many Listeria species cause invasive and noninvasive food-borne listeriosis (Zamuz et al., 2021). *Listeria monocytogenes* can cause meningitis, encephalitis, miscarriage, stillbirth in pregnant women, and death.

Additionally, symptoms such as diarrhea, nausea, vomiting, muscle pains, and fever are observed (Moabelo et al. 2023). Thus, the current study was to investigate the incidence of bacterial contamination in imported chilled and frozen beef

2. MATERIAL AND METHODS

Ethical approval

Following the approval of the research proposal by the Care and Use Committee Research Ethics, Faculty of Veterinary Medicine, Benha University (BUC-VTM-Oc-02-24), Egypt, the study was carried out.

2.1. Samples Collection

One hundred of imported chilled and frozen beef samples (50 of each) were randomly selected from supermarkets in Cairo and Qalyubia governorates. The samples were promptly transported to the laboratory in an icebox and

packaged in a sterile plastic bag. All samples underwent bacteriological analysis.

2.2. Sample Preparation

In brief, the beef samples were prepared as described in (ISO, 2017a). In a septic manner, a 25 g meat sample was homogenized with 225 ml of 0.1 % peptone water in a sterile stomacher bag for 2 minutes using a Stomacher (400R, Seward, UK). From the homogenized sample, serial decimal dilutions were made, and 100 µL was placed on agar plates and tested for Aerobic Mesophilic Bacteria (AMB), *Staphylococcus aureus*, isolate salmonella, *Escherichia coli,* and *Listeria monocytogenes*.

2.3. Bacteriological analysis of meat samples

2.3.1. Aerobic mesophilic bacteria (AMB) enumeration

AMB counts were determined by plating 100 µL of beef homogenate onto standard plate count agar using the spread plate method. Plates incubated at 37 ◦C for 24 hours under aerobic conditions (ISO, 2013).

2.3.2. Staphylococcus aureus enumeration

Staphylococcus aureus Enumeration was conducted in accordance with (ISO, 2021) using Baird-Parker (BP) agar medium (Oxoid, UK) with the addition of egg yolk tellurite emulsion (Oxoid, UK). BP dry surface plates were surface-plated with 0.1 mL of the appropriate homogenate dilutions. Inoculated plates were incubated at 37°C for 24-48 hours. Black colonies with an opaque halo on BP agar are likely *S. aureus*. Each plate's numbers were expressed as cfu/g.

2.3.2.1. Staphylococcal enterotoxins (SEs)

SEs were detected and typed using (Klotz et al., 2003) procedures with slight modification. For 18–24 hours, *S. aureus* isolates were cultured in tryptone soy broth at 37°C with shaking. After growth, the culture supernatant was centrifuged at $900 \times g$ for 20 minutes to detect enterotoxins. The commercially available SET-RPLA detected enterotoxins according to manufacturer directions. In summary, supernatant-diluted antisera-sensitive latex reagents are incubated overnight.

2.3.3. Isolation and identification of some foodborne pathogens.

2.3.3.1. Isolation and identification of Salmonella spp.

Salmonella was identified in accordance with (ISO, 2017b) by adding 1 ml from each sample to 9 ml of Rappaport-Vassiliadis broth (Oxoid). A loopful of Rappaport-Vassiliadis broth was spread onto Oxoid xylose lysine deoxycholate (XLD) agar and incubated at 37 °C for 18–24 hours after an overnight aerobic incubation. API 20E (bioMérieux, Marcy-l'Étoile, France) identified salmonella biochemical colonies. The Kaufman-White technique was used to serotype all biochemically validated Salmonella isolates using commercial antisera (SIFIN in Berlin,

Germany) targeting the somatic (O) and flagellar (H) antigens (Popoff et al., 2004).

3.3.2. Isolation and identification of E. coli

E. coli was isolated according to protocol (ISO, 2018). Briefly, from previously prepared meat samples, A sterile pipette was used to transfer 1 mL of diluted meat samples into a test tube with nutritional broth after pre-enrichment and incubate overnight at 37 °C. A loop of overnight culture was streaked onto Eosin Methylene Blue agar in duplicate and incubated at 37°C for 18-24hr. Three presumptive *E. coli* from each selective agar plate were picked and then subcultured to obtain a pure culture*,* and identification was performed using standard biochemical procedures including Gram,s staining, catalase, oxidase, indole, methyl red, Voges–Proskauer tests, and a sugar fermentation test using triple sugar iron agar. Positive isolates were stored in nutrient broth containing 50% (v/v) glycerol at 20°C for further study.

3.3.3 Isolation and identification of Listeria monocytogenes

To isolate *Listeria monocytogenes*, one ml from a previously prepared meat sample was added to 9 ml of half Fraser broth (Oxoid, England), homogenized, and incubated aerobically at 30°C for 24±2 hours (ISO, 2017c). Transfer 0.1ml of primary enrichment culture to 10ml Fraser broth (Oxoid, England) and incubate at 35°C or 37°C for 48±2 hours. A loopful of incubated Fraser broth was smeared onto PALCAM (Oxoid, England) agar plates and incubated at 37°C for 24±3 hours. The bacterial morphological and biochemical properties were assessed through the utilization of Gram,s staining, the catalase test, the sugar fermentation test, and the motility test, in accordance with the FDA BAM and ISO 11290 methods (Scotter, 2001)**.**

4. Statistical Analysis

Statistical analysis was performed on the collected data using One-Way ANOVA (Analysis of Variance) test, SPSS version 22.0 (IBM Corp., Armonk, N.Y., USA). The statistical significance was set at $P < 0.05$.

3. RESULTS

The present study reveals a statistically significant difference in aerobic mesophilic bacterial counts (AMB) between imported chilled and frozen beef samples ($P < 0.05$). The AMB counts for imported chilled beef samples varied between 5.3×10^4 and 4.7×10^6 cfu/g, with an average of $8.35\times10^5\pm1.06$cfu/g. In contrast, the imported frozen samples that were examined exhibited a mean value of $3.11\times10^5\pm0.29$cfu/g, as data is presented in Table 1. In addition, eleven out of fifty samples of chilled meat (22%) and 14% of the frozen beef samples (7 out of 50) were found to be unsatisfactory based on the APC limit specified by EOS 3602/2013 and EOS 1522/2018, respectively.

Table 1 Aerobic mesophilic bacteria (AMB) counts (cfu/g) in imported chilled and frozen beef.

Meat samples	AMB			Acceptability	
				Accepted	Unaccepted
	Min	Max	Mean ± S.E*	No. (%)	No. (%)
Chilled meat	5.3×10^4	4.7×10^6	$8.35\times10^5\pm1.06^a$	39 (78)	11(22)
Frozen meat	9.7×10^3	1.5×10^6	$3.11\times10^5\pm0.29^b$	43 (86)	7(14)

S.E:Standard error of mean
Maximum permissible limit (MPL)according to (Egyptian Organization for Standardization and Quality (EOS) 3602/2013) for chilled beef and (EOS1522/2018 2018) for frozen beef) for AMB = $> 10^6$

The mean counts of *Staphylococcus aureus* for imported chilled beef were $1.48\times10^3\pm 0.15$cfu/g whereas for imported

frozen meat were $8.75\times10^2\pm 0.94$cfu/gas illustrated in Table 2. Moreover, in compliance to the thresholds established by

EOS (1522/2018) for frozen meat and (3602/2013) for chilled meat, *Staphylococcus aureus* levels were deemed

unacceptable in eight of fifty frozen beef samples (16 %) and twelve of fifty chilled meat samples (24 %).

Table 2 *Staphylococcus aureus* counts (cfu/g) and acceptability levels in imported chilled and frozen beef.

Meat samples	Chilled meat	Frozen meat
Enterotoxins	No. (%)	No. (%)
SEA	1 (2)	-
SED	1(2)	1(2)
SEA+SEC	-	1(2)
SEC+SED	1(2)	-
Total	3(6)	2(4)

S.E: Standard error of mean, MPL for *Staphylococcus aureus* counts = 10^2

Table 3 shows the enterotoxin distribution in *Staphylococcus aureus* strains from imported chilled and frozen beef samples. Only 6% of chilled beef samples were positive for staphylococcal enterotoxins with 2% for

staphylococcal type A (SEA), 2% for SED, and 25 for SEC+SED, in contrast, 4% of imported frozen beef samples tested positive for staphylococcal enterotoxins with 2% for SED, and 2 % for SEA+SEC.

Table 3 Prevalence of Staphylococcus enterotoxins in imported chilled and frozen beef.

Meat samples	Chilled meat	Frozen meat
Salmonellae spp	No. (%)	No. (%)
S. Enteritidis	2 (4)	1 (2)
S. Haifa	-	1(2)
S. Montevideo	1 (2)	-
S. Shangani	1 (2)	-
S. Takoradi	-	1(2)
S. Typhimurium	1(2)	-
Total	5 (10)	3(6)

Table 4 reveals that *S. Enteritidis* (4 %), *S. Montevideo* (2 %), *S. Shangani* (2 %), and *S. Typhimurium* (2 %) were found in imported chilled beef, whereas *S. Enteritidis* (2%),

S. Haifa (2 %), and *S. Takoradi* (2 %) were found in imported frozen beef samples.

Table 4 Prevalence of salmonella spp. in imported chilled and frozen beef samples.

Meat samples	*Staphylococcus aureus*			Acceptability	
				Accepted	Unaccepted
	Min	Max	Mean ± S.E	No. (%)	No. (%)
Chilled meat	<10^2	5×10^3	$1.48\times10^3 \pm 0.15^a$	38 (76)	12 (24)
Frozen meat	<10^2	2×10^3	$8.75\times10^2 \pm 0.94^b$	42 (84)	8 (16)

Table 5 presents the serotyping results for *E. coli* isolated from the examined imported chilled beef (7 serotyping) and imported frozen beef (4 serotyping). Specifically, the following *E. coli* strains were detected in chilled meat samples: *E.coli* O_{17}:H_{18} (2 %), *E.coli* O_{111}:H_2 (2 %), and

E.coli O_{124} (2 %); in imported frozen meat samples, the following strains were detected: *E.coli* O_{26}:H_{11} (4 %), *E.coli* O_{55}:H_7 (2 %), *E.coli* O_{91}:H_{21} (2 %), and *E.coli* O_{119}:H_6 (2 %); however, *E.coli* O_{128}:H_2 failed to be detected.

Table 5 Prevalence of *E. coli* in imported chilled and frozen beef samples.

Meat samples	Chilled meat	Frozen meat	Strain type
E. coli strains	No. (%)	No. (%)	
O_{17}: H_{18}	-	1(2)	EPEC
O_{26}: H_{11}	2 (4)	-	EHEC
O_{55}: H_7	1(2)	-	EPEC
O_{91}: H_{21}	1(2)	-	EHEC
O_{111}: H_2	-	1(2)	EHEC
O_{119}: H_6	1(2)	-	EPEC
O_{124}	-	1(2)	EIEC
O_{128}: H_2	2(4)	1(2)	ETEC
Total	7(14)	4 (8)	

EPEC = Enteropathogenic *E. coli*, ETEC = Enterotoxigenic *E. coli*, EIEC = Enteroinvasive *E. coli*, EHEC = Enterohaemorrhagic *E. coli*

E. coli and salmonella-free food is mandated by international organizations' food safety standards (EOS No. 3602/2013 and 1522/2018). Seven samples of imported chilled meat (14%) and four samples of imported frozen meat (8%) were rejected because of *E. coli* in this study. Salmonella spp. contamination led to the rejection of five (10%) samples of imported chilled meat and three (6%) samples of imported

frozen meat. In compliance with the *L. monocytogenes* standards set by the (EOS 3602/2013) for chilled meat and the (EOS 1522/2018) for frozen meat, both of which are devoid of contamination, the percentage of chilled meat that was considered unacceptable was 6% (3 samples) and 1 % (one sample), respectively, rendering them hazardous for human consumption (Table 6).

Table 6 Acceptability of chilled and frozen beef samples based on their contamination with *E. coli*, Salmonella spp., and *Listeria monocytogenes*.

strains	*E.E. coli*		Salmonella spp.		*Listeria monocytogens*	
Meat samples	Accepted	Unaccepted	Accepted	Unaccepted	Accepted	Unaccepted
	No. (%)	No. (%)	No. (%)	No. (%)	No. (%)	No. (%)
Chilled	43(86)	7(14)	45 (90)	5(10)	47(94)	3(6)
Frozen	46 (92)	4 (8)	47(94)	3(6)	49(98)	1(2)
Total (100)	89 (89)	11(11)	92(92)	8(8)	96(96)	4(4)

MPL for (*E.coli, salmonella spp. And Listeria monocytogenes*)= Free according EOS.

4- DISCUSSION

Microbiological foodborne hazards have attracted the attention of the food safety management system. AMB counts were significantly higher in imported chilled beef than imported frozen samples. Similar results have been reported by (Hassanien et al., 2020). Both chilled and frozen beef samples were rejected by EOS at 22% and 14%, respectively. Equipment and cleanliness affect aerobic bacterial load. Increased aerobic mesophilic counts may suggest poor processing cleanliness (Ma et al., 2014). The elevated levels of AMB may not directly indicate a concern to human health but can serve as markers of hygienic quality in food processing areas and products (Rodríguez-Melcón et al., 2022). *Staphylococcus aureus* causes numerous clinical illnesses worldwide, and its link with food poisoning has garnered public attention. The current investigation found that compared to frozen meat samples, chilled meat had significantly higher levels of *S. aureus*. According to EOS standards, 24% of chilled and 16% of frozen meat samples had unsatisfactory *Staphylococcus aureus* levels. In contrast, earlier investigations found *S. aureus* positivity in raw red meat at 29.4 % in Algeria (Chaalal et al., 2018) and 26.31 % in Iran (Safarpoor Dehkordi et al., 2017). *Staphylococcus aureus* is usually connected with skin and clothing; however, it can also be caused by dirty food processing practices such using soiled cutting boards, blades, and butcher shop staff' infamously poor hygiene (Gebeyehu, 2013). *S. aureus* and other pathogens can contaminate meat due to poor slaughter cleanliness and other abattoir errors such as faulty evisceration, which increases the risk of gut infections (Jaja et al., 2020). Additionally, in chilled meat, staphylococcal enterotoxins (SEs) were higher (6%) than in frozen meat (4%). These Preformed toxins cause nausea, vomiting, abdominal pain, and diarrhea, mimicking staphylococcal food poisoning. Two to six hours are typical for incubation. Diarrhoea usually follows rapid vomiting (Balaban and Rasooly, 2000). Furthermore, *E. coli* was detected in 8% of chilled and 12% of frozen retail red meat, respectively. Consistent with previous research, Gamal et al. (2020) found *E. coli* $O_{55}:H_7$ in one of the fresh meat samples tested. in contrast, Ahmed & Shimamoto (2014) found *E. coli* $O_{157}:H_7$ in 3.4% of samples and found it more prevalent in dairy than meat. Pathogenic *E. coli* infection can cause chronic diarrheal illness, vomiting, and more serious medical disorders such as traveler's diarrhea and hemolytic uremic syndrome (HUS). Salmonella is the most prevalent causative agent of foodborne diseases on a global scale. In a comparable trend, the prevalence of salmonella spp. in chilled meat (10 %) was greater than in frozen meat (6 %). Consistent with these results, Hendriksen et al. (2011) reported that *S. Enteritidis* and *S. Typhimurium* are the most common serovars in meat and animal products and cause most human diseases. Salmonellosis is characterized by fever, diarrhea, and severe cramps, and can develop up to 72 hours after ingestion (Antunes et al., 2016). The present investigation found that *L. monocytogenes* caused the rejection of 3 (6%) of imported chilled meat and 1 (2 %) of imported frozen meat. A nearly similar result was reported

by Ismaiel et al. (2013) who found 3.33 % *L. monocytogenes* in frozen lean beef, although the high values was 7.2 % (Liu et al., 2020). Foodborne pathogen *L. monocytogenes* can cause moderate gastroenteritis to invasive listeriosis (Horita et al., 2018). Certain food products have consistently shown increased vulnerability to contamination by *L. monocytogenes* due to the bacterium's capacity to multiply in cold temperatures, in addition, the *L. monocytogenes* is capable of surviving at temperatures below freezing and can thrive within a temperature range of 1°C to 45°C (Saraiva et al. 2016).

5. CONCLUSIONS

The present data indicated that imported chilled beef samples had more Aerobic mesophilic bacteria, *S. aureus*, than imported frozen meat samples. Furthermore, higher rates of salmonella, *E. coli*, and *Listeria monocytogenes* in imported chilled beef. According to this study, chilled beef is a public health risk. The results from this study emphasize how crucial it is to maintain proper hygiene protocols during meat processing and retail environments. It is essential to establish programs that help prevent contamination or inhibit the growth of bacteria in meat. Other important measures include maintaining an appropriate temperature, using good manufacturing practices, and ensuring proper cleaning, sanitation, and hygiene.

6. REFERENCES

1. Ahmed A.M. and Shimamoto T., 2014. Isolation and molecular characterization of *Salmonella enterica, Escherichia coli* O157:H7 and Shigella spp. from meat and dairy products in Egypt. International Journal of Food Microbiology 168–169, 57–62.
2. Antunes P., Mourão J., Campos J., Peixe L., 2016. Salmonellosis: the role of poultry meat. Clinical Microbiology and Infection 22, 110–121.
3. Balaban N. and Rasooly A. 2000. Staphylococcal enterotoxins. International Journal of Food Microbiology 61, 1–10.
4. Borch E. and Arinder P., 2002. Bacteriological safety issues in red meat and ready-to-eat meat products, as well as control measures. Meat Science 62, 381–390.
5. Chaalal W., Chaalal N., Bourafa N., 2018. Characterization of *Staphylococcus aureus* Isolated from Food Products in Western Algeria. Foodborne Pathogens and Disease 15, 353–360.
6. Egyptian Organization for Standardization and Quality (EOS) 2013. Reports related to No. 3602/2013. Chilled meats, Egyptian standards, Ministry of industry, Egypt.
7. Egyptian Organization for Standardization and Quality (EOS) 2018. Reports related to No. 1522/2018 Frozen meats, Egyptian standards, Ministry of industry, Egypt.
8. Ehuwa O., Jaiswal A.K., Jaiswal S., 2021. Salmonella, Food Safety and Food Handling Practices. Foods 10, 907.
9. Gamal N., Abd El-Tawab A., Elhofy F., Maarouf A., 2020. Phenotypic characterization of some food poisoning bacteria isolated from meat and meat products in Kaliobia, Egypt. Benha Veterinary Medical Journal 38, 146–151.
10. Gebeyehu A., 2013. Evaluation of Microbial Load of Beef of Arsi Cattle in Adama Town, Oromia, Ethiopia. Journal of Food Processing & Technology 4, 6.

11. Hassanien F., shaltout F., Fahmy M., Elsukkary H., 2020. Bacteriological quality guides in local and imported beef and their relation to public health. Benha Veterinary Medical Journal 39, 125–129.

12. Hendriksen R.S., Vieira A.R., Karlsmose S., 2011. Global Monitoring of Salmonella Serovar Distribution from the World Health Organization Global Foodborne Infections Network Country Data Bank: Results of Quality Assured Laboratories from 2001 to 2007. Foodborne Pathogens and Disease 8, 887–900.

13. Horita C.N., Baptista R.C., Caturla M.Y.R., 2018. Combining reformulation, active packaging and non-thermal post-packaging decontamination technologies to increase the microbiological quality and safety of cooked ready-to-eat meat products. Trends in Food Science & Technology 72, 45–61.

14. Ismaiel A.A.R, Ali A.E.S., Enan G., 2013. Incidence of Listeria in Egyptian meat and dairy samples. Food Science and Biotechnology 23, 179–185.

15. ISO 11290-1, 2017c. Microbiology of the food chain Horizontal method for the detection and enumeration of Listeria monocytogenes and of Listeria spp. Part 1: Detection method. International Organization for Standardization, Geneva.

16. ISO 16649-1, 2018. Microbiology of the food chain — Horizontal method for the enumeration of beta-glucuronidase-positive Escherichia coli — Part 1: Colony-count technique at 44 degrees C using membranes and 5-bromo-4-chloro-3-indolyl beta-D-glucuronide. International Organization for Standardization, Geneva.

17. ISO 4833-1, 2013. Microbiology of the food chain Horizontal method for the enumeration of microorganisms Part 1: Colony count at 30 °C by the pour plate technique. International Organization for Standardization, Geneva.

18. ISO 6579-1, 2017b. Microbiology of the food chain Horizontal method for the detection, enumeration and serotyping of Salmonella Part 1: Detection of Salmonella spp. International Organization for Standardization, Geneva.

19. ISO 6887-2, 2017a. Microbiology of the food chain. Preparation of test samples, initial suspension and decimal dilutions for microbiological examination. International Organization for Standardization, Geneva.

20. ISO 6888-1, 2021. Microbiology of food and animal feeding stuffs. Horizontal method for the enumeration of coagulase-positive staphylococci (*Staphylococcus aureus* and other species). International Organization for Standardization, Geneva.

21. Jaja I.F., Jaja C.J.I., Chigor N.V., 2020. Antimicrobial Resistance Phenotype of *Staphylococcus aureus* and *Escherichia coli* Isolates Obtained from Meat in the Formal and Informal Sectors in South Africa. Biomed Research International 2020, 3979482.

22. Kebede M.T. and Getu A.A., 2023. Assessment of bacteriological quality and safety of raw meat at slaughterhouse and butchers' shop (retail outlets) in Assosa Town, Beneshangul Gumuz Regional State, Western Ethiopia. BMC Microbiology 23, 403.

23. Klotz M., Opper S., Heeg K., Zimmermann S., 2003. Detection of *Staphylococcus aureus* enterotoxins A to D by real-time fluorescence PCR assay. Journal of Clinical Microbiology 41, 4683–4687.

24. Leygonie C., Britz T.J., Hoffman L.C., 2012. Meat quality comparison between fresh and frozen/thawed ostrich M. iliofibularis. Meat Science 91, 364–368.

25. Liu Y., Sun W., Sun T., 2020. The prevalence of *Listeria monocytogenes* in meat products in China: A systematic literature review and novel meta-analysis approach. International Journal of Food Microbiology 312, 108358.

26. Ma F., Yao J., Xie T., 2014. Multispectral imaging for rapid and non-destructive determination of aerobic plate count (APC) in cooked pork sausages. Food Research International 62, 902–908.

27. Manges A.R. and Johnson J.R., 2012. Food-Borne Origins of *Escherichia coli* Causing Extraintestinal Infections. Clinical Infectious Diseases 55, 712–719.

28. Moabelo K.C., Gcebe N., Gana J., 2023. Contamination of beef and beef products by Listeria spp. and molecular characterization of *L. monocytogenes* in Mpumalanga, South Africa. Journal of Food Safety 43, 13055.

29. Mohammed H.H.H., He L., Nawaz A., Jin G., Huang X., Ma M., Khalifa I., 2021. Effect of frozen and refrozen storage of beef and chicken meats on inoculated microorganisms and meat quality. Meat Science 175, 108453.

30. Popoff M.Y., Bockemühl J., Gheesling L.L., 2004. Supplement 2002 (no. 46) to the Kauffmann–White scheme. Research in Microbiology 155, 568–570.

31. Rodríguez-Melcón C., Esteves A., Panera-Martínez S., 2022. Quantification of total and viable cells and determination of serogroups and antibiotic resistance patterns of *Listeria monocytogenes* in chicken meat from the North-Western Iberian Peninsula. Antibiotics 11, 1828.

32. Safarpoor Dehkordi F., Gandomi H., Basti A.A., 2017. Phenotypic and genotypic characterization of antibiotic resistance of methicillin-resistant *Staphylococcus aureus* isolated from hospital food. Antimicrobial Resistance and Infection Control 6, 1-11.

33. Saraiva C., Fontes M.C., Patarata L., 2016. Modelling the kinetics of *Listeria monocytogenes* in refrigerated fresh beef under different packaging atmospheres. LWT - Food Science and Technology 66, 664–671.

34. Scotter S. 2001. Validation of ISO method 11290 Part 2. Enumeration of *Listeria monocytogenes* in foods. International Journal of Food Microbiology 70,121–129.

35. Xu X., Rothrock M.J., Reeves J., 2022. Using E. coli population to predict foodborne pathogens in pastured poultry farms. Food Microbiology 108, 104092.

36. Zamuz S., Munekata P.E.S., Dzuvor C.K.O., 2021. The role of phenolic compounds against *Listeria monocytogenes* in food. A review. Trends in Food Science and Technology 110, 385–392.