

Review Article

A Review on the Microbial Surface Contaminants of the Animal Carcasses

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Abstract

The flesh of the slaughtered animals is believed to be sterile. However, after slaughter, meat of different animal species is exposed to a vast array of microorganisms during processing of the carcasses. Carcass processing includes slaughtering, dressing, evisceration, quartering, and further transportation, and distribution. Several sources of microbial contamination might contribute to the microbial status of the different carcasses including internal sources such as the ruminal content of the animal, blood, hide and skin, and external sources such as butchers hands, clothes, knives, cutting boards, and abattoir environment including walls, floors, washing water, etc. One major task of the food safety sector is to ensure the hygienic measures adopted during all steps of meat processing. Therefore, this study was undertaken to review the microbial surface contaminants of the animal carcasses. This review concluded that efficient hygienic measures should be adopted during all steps of handling of the meat in order to obtain a high keeping quality meat with low initial microbial load.

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INTRODUCTION

Meat is a rich source of highly bioactive compounds including essential amino acids, vitamins such as Vitamin B complex, minerals and trace elements such as zinc, iron, and copper (Sallam and Morshdy, 2008; Elabbasy *et al.*, 2021). Moreover, Cattle and buffalo carcasses contain considerable concentrations of carotenoids and retinol (Darwish *et al.*, 2016).

Prior to slaughter, healthy animal skeletal muscle has traditionally been thought to be sterile, with the exception of the lymph nodes (Darwish *et al.*, 2018). Although intrinsic bacteria may occasionally exist in muscle tissues at low levels, this is not the most frequent cause of contamination. By far, extrinsic causes are the main cause of meat and carcass contamination. During steps in the harvest stage where the external surfaces of the carcass are exposed to potential sources of contamination, bacteria may come into contact with the beef carcass. According to Lahr (1996), feces, ruminal contents, and the hide can contaminate the carcasses microbiologically. Additional cross-contamination points in the slaughter process include carcass-to-carcass contact, human touch, structural elements of the facility, and processing tools and equipment. Fortunately, most of the microflora that is transferred to carcass surfaces is not pathogenic, despite being unsightly (Huffman, 2002).

A vast array of microorganisms can invade the meat surfaces including *Staphylococcus aureus* (*S. aureus*), *E. coli*, *Salmonella* spp., *Pseudomonas*, *Acinetobacter/Moraxella*, *Aeromonas*, *Alteromonas putrefaciens*, *Lactobacillus*, and *Brochothrix thermosphac-*

ta. The pathogenic bacteria of most concern include *Escherichia coli* O157:H7, *Salmonella* spp., *Listeria monocytogenes*, *Campylobacter*, *Clostridium botulinum*, *Clostridium perfringens*, *S. aureus*, *Aeromonas hydrophila*, and *Bacillus cereus* (Darwish *et al.*, 2022; Darwish *et al.*, 2023).

The quality and safety of the global food supply are of utmost importance. According to the Centers for Disease Control and Prevention (CDC, USA), 3000 people die from food-borne illnesses each year, 128 000 are hospitalized, and one in six Americans (about 48 million people) get sick (CDC, 2018). 43 400 cases of food- and water-borne outbreaks were reported by the member states of the European Union (EU) in 2017. According to the European Food Safety Authority and the European Centre for Disease Prevention & Control (EFSA-ECDC, 2018); 642 of the outbreaks were classified as "strong-evidence" food-borne outbreaks, with 385 (or 60%) of them attributable to foods of animal origin, with meat and meat products being the most frequently involved food. Food-borne outbreaks continue to occur despite numerous advancements in food biotechnology, contemporary methods, and the application of the Hazard Analysis Critical Control Points management system (HACCP) (U.S. Food & Drug Administration, 2018a; 2018b; Nielsen *et al.*, 2021).

As a major task of the food safety sector it is necessary to investigate the microbiological quality of the meat and the potential sources of the microbial contamination of the carcasses. Therefore, in this review, we would like to introduce a review on the microbial surface contaminants of the bovine carcasses, their contact surfaces, and other meat products.

Microbial contamination of meat and their contact surfaces

It is important to keep an eye on animal products to make sure that humans can still eat safe meat. Biological, physical, and chemical risks are possible in beef meat at every stage of the supply chain, from slaughter to consumption. Healthy cattle typically have pathogenic bacteria in their digestive tracts. These microorganisms can also be found on the hides of living animals that have been contaminated by feces. During slaughtering, particularly when done on the ground without a carcass suspension system and with careless evisceration that spreads intestinal content onto the meat surface, these microorganisms can then be transferred to the surface of previously sterile meat. During the slaughter process, contact with the animal's skin and hair, limbs, blood, stomach, and gut can infect the carcasses of cattle (Darwish *et al.*, 2022).

Guerrero and Taylor (1994) reported that when carcasses are stored in semitropical climes or exposed to high temperatures in industrialized areas, meat surface contamination by microbes has been a significant issue.

New meat and poultry inspection laws being adopted in the United States are proof that attempts to limit contamination of raw meat have escalated as a result of increased consumer awareness and concern about microbiological foodborne infections. The new regulations have established microbiological testing criteria for *Escherichia coli* and *Salmonella*, as a means of evaluating plant performance, in addition to requiring operation of meat and poultry slaughtering and processing plants under the principles of the hazard analysis critical control point (HACCP) system. The development and commercial use of meat and poultry decontamination techniques have received renewed and increased interest as a result of these advancements. Live animal cleaning and washing, chemical de-hairing, knife-trimming of carcasses to remove physical contaminants, steam/hot water vacuuming for spot-cleaning/decontamination of carcasses, spray washing/rinsing of carcasses with water of low or high pressures and temperatures or chemical solutions, and exposure of carcass sides to pressurized steam are some of the technologies developed and evaluated for decontamination. Under the right circumstances, the technologies used on carcasses may lower mean microbiological counts by one to three log colony forming units (cfu)/cm², and some of them (such as steam-vacuuming, carcass spray-washing with water, chlorine, organic acid, or trisodium phosphate solutions, hot water deluging/spraying/rinsing, and pressurized steam) have been approved and are used in commercial applications.

Gibbons *et al.* (2006) mentioned that in addition to determining the incidence of *Listeria spp.*, *Salmonella spp.*, *Escherichia coli*, and *Campylobacter spp.* in the meat products and air inside the meat processing plant, the investigation was carried out to look into the potential source(s) of *Listeria* contamination of recalled meat products. After the plant had been thoroughly cleaned and shut down, surfaces that came into touch with food as well as raw and processed meat products were further tested for the presence of *Salmonella spp.*, *Listeria spp.*, and *Campylobacter spp.* *Salmonella*, *Listeria*, and *Campylobacter* species were examined in feces and effluent samples taken from a piggery close to the factory. The studied organisms were absent from air samples and surfaces that came into contact with the examined samples. *Campylobacter coli* was detected in ten (58.8%) of the 17 effluent samples and four (11.8%) of the 34 fecal samples. 10 (90.9%) of the 11 raw meat products tested were found to have either one or both of the *Listeria spp.* or *E. coli*. Of the 32 processed meat prod-

ucts examined, 11 (34.4%) tested positive for either one or more of the following pathogens: *E. coli*, *Salmonella spp.*, *Listeria spp.*, and *Campylobacter spp.* According to international regulations, 11 (61.1%) of 18 processed meat products had levels of aerobic bacteria that were too high. According to the obtained results, *Listeria spp.* and other pathogens were less likely to spread when better hygiene was practiced when handling items and on surfaces that came into contact with meat products. Therefore, it may be deduced that the issue at the facility is caused by shortcomings in proper sanitary practices, insufficient heat treatments, or the presence of pathogens, mainly *Listeria*, in biofilms on various surfaces that occasionally or continuously contaminate completed meat products.

Facilities used for slaughter at various stages of meat processing, such as skinning, evisceration, storage, and distribution, may contaminate meat and meat products. Traditional ways of handling, processing, and selling meat weaken quality in the majority of developing nations, where poor sanitation causes significant product loss and increases the risk of food-borne illnesses. In most developing nations, exterior meat contamination is a substantial issue, and carcass surface microbial infection has been repeatedly shown to have a considerable impact on the meat shelf life. Fecal matter is a significant source of microbial contamination and may come into contact with carcasses directly or indirectly through polluted and filthy personnel, surfaces, equipment, and other factors as illustrated in Sudan (Abdalla *et al.*, 2009).

Ali *et al.* (2010) determined how frequently meat sold in Karachi, Pakistan, was contaminated. Microbiological contamination was examined in 250 raw meat samples and 90 surface swabs from the environment and meat processing equipment. 340 samples were examined, and it was discovered that 84% of them contained germs, including *Klebsiella*, *Enterobacter*, *S. aureus*, and *Bacillus subtilis*. Of the bacterial isolates, 550 (or 66%) had the potential to be pathogens. Of them, 208 and 342 isolates, respectively, were from environmental and meat samples. *Escherichia coli* O157:H7, *Listeria*, *Salmonella* Enteritidis, and *Shigella* species were among the food-borne pathogens isolated from meat samples, whereas *S. aureus* and *Shigella* species were found in ambient samples. Additionally, four *Brucella* species strains were found in samples of meat. The range of the total aerobic counts was 10⁸–10¹⁰ CFU/g or cm². It was noted that there was resistance to a variety of antibiotics. A typical range of 62 to 75% of people were resistant to ampicillin, amoxicillin, novobiocin, and cefaclor. A third of *Salmonella* isolates exhibited ampicillin resistance. No evidence of quinolone resistance was found. 88 (16%) harmful bacteria, including *E. coli*, *Klebsiella*, *Enterobacter* species, and *S. aureus*, formed biofilms.

The presence of *Listeria monocytogenes*, *Salmonella spp.*, *Bacillus cereus*, *Staphylococcus spp.*, *Enterococcus spp.*, and *Escherichia coli* was examined in 2005–2006 in three dairy cattle farms (120 samples), one dairy (124 samples), and two meat processing plants (160 samples) as well as in raw food materials, food products, and on surfaces in contact with food after sanitation. There were 1409 different isolates in total. On a few isolates, the epidemiological characterization, virulence factor identification, and antibiotic resistance testing were done. During the production process, the amount of bacterial contamination generally reduced (the contamination of food items was lower than that of raw materials). Even after sanitation, the level of contamination on surfaces that came into touch with food was still quite significant. Additionally, particular microbiological profiles were discovered on the inside surfaces of equipment in dairy operations, where it is possible to find staphylococci-like genetically

closely related multi-resistant strains persisting in biofilm communities. Even though the frequency of potentially dangerous pathogens was low, the meat-processing facilities were primarily contaminated by bacteria such *L. monocytogenes*, *Salmonella* spp., and *E. coli* that was shiga-toxin positive. The majority of *B. cereus* isolates, of which 76% were positive for diarrhogenic enterotoxin, were found on the interior surfaces of equipment and in heat-treated goods.

In Ethiopia, Gurmu and Gebretinsae (2013) examined the pathogens and indicator microorganisms for cleanliness in touch surfaces, minced meat, and butcher shops in Addis Abeba, Ethiopia. In addition, a checklist was utilized to measure the cleanliness of the venues, and a questionnaire/checklist was employed to gauge the food handlers' familiarity with food safety. According to this study, the average microbial counts of minced meat and contact surface materials in butcher shops ranged from 2.35 to 6.50 log cfu/g and from 1.80 to 6.30 log cfu/cm², respectively, for total aerobic mesophilic, staphylococci, *Enterobacteriaceae*, total coliforms, and aerobic spores and yeasts/molds. There were statistically significant changes between the mean microbial counts of minced beef samples collected in the morning and the afternoon. In contact surface and minced meat samples, the proportions of *E. coli*, *Salmonella*, and *S. aureus* were 43.75 and 29.17%, 6.25 and 4.17%, and 37.50 and 37.50%, respectively. According to the study, poor personal and workplace hygiene was associated with greater microbial loads in minced beef samples and contact surface materials. Significant contributing causes for the contamination of beef have also been identified as being inadequate training for food handlers in butcher shops and a broken cold chain. Moreover, Tegegne and Phyto (2017) conducted a study to evaluate the sanitary standards and microbiological quality of meat contact surfaces at slaughterhouses and retail establishments in Jijjiga town, Ethiopia. A total of ninety pooled swab samples were taken from abattoir floor surface, butchers' hands, hooks and knives and cutting boards to assess the presence and load of *S. aureus*, *Escherichia coli* O157:H7, *Listeria monocytogenes*, aerobic bacteria (aerobic plate counts or APCs), fecal coliforms (FCs), yeast and molds, and *Campylobacter* spp. According to the data, butchers' hands at retail stores (6.43±0.34 cfu/cm²) and abattoirs (6.03±0.03 cfu/cm²) had the highest average *S. aureus* and *E. coli* O157:H7 counts, respectively. Only the floor surface of the abattoir had *Campylobacter* species. *L. monocytogenes* was not found in any of the samples from surfaces that came into contact with meat, making up 3.33% of the total samples that tested positive for *Campylobacter* spp. The abattoir floor surface had the greatest counts of FCs (6.25±0.075 log₁₀ cfu/cm²) and yeast and molds (5.19±0.513 log₁₀ cfu/cm²), whereas the butchers' hands had the highest APC counts (6.08±0.126 log₁₀ cfu/cm²). This finding suggests that retail meat touch surfaces and slaughterhouse surfaces could both be sources of meat contamination. In order to improve the overall microbiological quality and sanitary level of meat contact surfaces and protect the customer from foodborne pathogens, effective hygiene practices should be implemented.

Stellato et al. (2016) studied the differences between small-scale retail distribution (SD) and large-scale retail distribution (LD) facilities, as well as the impact of the meat processing environment on the initial fresh meat infection. Over the course of two sampling campaigns, samples were gathered from butcheries (n = 20), including LD (n = 10) and SD (n = 10) facilities. Fresh pieces of beef and pig as well as swabs from the butcher's hand, the knife, and the cutting board were included in the samples. More than 800 operational taxonomic units (OTUs) collapsed at the species level were present in the microbiota of both

ambient swabs and meat samples, demonstrating their extreme complexity. The 16S rRNA sequencing analysis revealed that the core microbiota, which included *Pseudomonas* spp., *Streptococcus* spp., *Brochothrix* spp., *Psychrobacter* spp., and *Acinetobacter* spp., were shared by 80% of the samples. In particular, meat samples had considerably greater levels of *Pseudomonas* and various *Enterobacteriaceae* members, whereas ambient swab samples had higher concentrations of *Brochothrix*, *Staphylococcus*, lactic acid bacteria, and *Psychrobacter*. When metabolic activities were taken into account by a predictive metagenomic analysis of the data, consistent grouping was also seen. Increases in pathways associated to amino acid and lipid metabolism were anticipated for the meat samples and were positively correlated with Proteobacteria, whereas increases in carbohydrate metabolism were predicted for the ambient swabs and were consistently linked to Firmicutes.

Diyantoro and Wardhana (2019) reported that to obtain safe, healthy, wholesome, and halal beef, it is expected that the supply of beef meat would not exceed the maximum microbiological contamination level. Concerns for food safety and shelf life in the production of meat arise from bacterial contamination during the slaughtering process. The purpose of this study was to evaluate the importance of microbial contamination and its potential hazards during the abattoir slaughtering process in ten Indonesian abattoirs. This study used visual observation, questionnaires, lab tests for bacterial contamination, and visual inspection alone. According to the findings, carcass cutting, which was not done by the abattoir (mean: 0.46 × 10⁶ CFU/g), had a substantial impact on the total plate count (TPC). Blood clearance on the floor position (mean: 40.34 × 10⁶ CFU/g) was the factor that had the biggest impact on the MPN of *Escherichia coli*. Blood removal from the floor position (mean: 52.88 × 10⁶ CFU/g) and carcass cutting performed outside of an abattoir (mean: 66.42 × 10⁶ CFU/g) had a substantial impact on *S. aureus* contamination.

Zadravec et al. (2020) studied surface molds that were growing on Croatian prosciutto and fermented sausages manufactured in various climate zones using various technologies (n = 160), and to link their existence to the levels of aflatoxin B1 (AFB1) and ochratoxin A (OTA). According to the findings, *Penicillium* (79%) was found to be the most prevalent contaminating mould, while *Aspergillus* (11%), *Eurotium* (7%) and *Mucor* (4%) species were found in significantly fewer isolates. Prosciuttos also showed a higher prevalence and greater diversity of contaminating mould than sausages, depending on the method of production and local climate. While AFB1 concentrations were only marginally higher than, OTA contamination (14% of samples) was substantially more common than that with AFB1 (8% of samples), with OTA concentrations reaching a maximum of 6.86 µg/kg. Some samples showed the presence of AFB1 despite the absence of toxicogenic molds, which can be attributable to tainted spices used in the manufacturing of TMP or indirect contamination through a carry-over effect.

CONCLUSION

This review indicated that meat can be contaminated with a wide range of microorganisms via external sources such as hides, skin, butcher hands, equipment and abattoir environment. Besides, internal sources such as the ruminal contents, fecal matter, and the animal ingesta. Therefore, strict hygienic measures should be adopted during all steps of meat processing.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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