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Review

Contribution of the Use of Microbiologically Contaminated Water in Slaughterhouses to Food Safety Risks

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ABSTRACT

Food animals can become contaminated with enteric pathogens during slaughter, with potentially significant consequences for food and public health safety. This overview examines the scientific evidence implicating slaughterhouses as critical points for microbiological contamination of meat, with particular focus on the role of process water as a potential source and vehicle for foodborne pathogens. Despite the extensive use of water—e.g., for carcass washing, equipment cleaning, and general hygiene, there is a notable lack of empirical data on how waterborne pathogens may contribute to food safety risks. This gap highlights the need to better characterize the potential for process water to act as a reservoir and a vehicle for foodborne pathogens. To address this, this overview proposes a framework for tracing, characterizing, and quantifying the food safety risks associated with water used during slaughter. It emphasizes the importance of generating experimental data on the survival, persistence, and fate of water-origin pathogens on or within meat. The ability of a pathogen to persist throughout processing and storage significantly influences its impact as a foodborne hazard, as it increases the likelihood that it reaches consumers at infectious doses. Establishing genetic relatedness among isolates recovered from slaughterhouse water, contaminated meat, and clinical cases of foodborne illness can confirm water as a contamination source. However, in general, robust microbiological and food chain surveys are needed to establish clearer links between water-mediated contamination in slaughterhouses and subsequent human illness. Addressing these research gaps is critical for designing effective interventions and ensuring meat safety from slaughter through distribution.

Abattoirs worldwide commonly wash carcasses with water to remove visible contaminants such as bone dust, blood clots, soil, and other debris (Bolton et al., 2001; Buncic & Sofos, 2012; Zweifel et al., 2015; Althaus et al., 2017; da Silva & Farag, 2021) or to reduce microbial contamination (Kochevar et al., 1997). Depending on the region, other commonly used water-based physical and chemical meat decontamination treatments include hot water or steam treatment, chemical solutions, electrolyzed or ozonated water, among others (Loretz et al., 2010; Buncic & Sofos, 2012). In most industrial slaughterhouses around the world, at several points along the slaughter line, carcasses, but equipment as well, may be sprayed with, rinsed or otherwise treated with or in water (Peyrat et al., 2008; Hernández et al., 2013; Schäfer et al., 2017; Rasschaert et al., 2020). Water, though vital for slaughterhouse operations, is however a potential vehicle of pathogens to meat carcass surfaces. It can also promote cross−contamination during slaughtering activities (Perez-Arnedo et al., 2021).

Studies have acknowledged that there are several complex contamination routes for meat in slaughterhouses, with experimental efforts characterizing or attempting to trace contamination along various steps, including scalding, evisceration, or postevisceration and water chilling, among other steps (Spescha et al., 2006; Wheatley et al., 2014; Shang et al., 2019). Process water used in slaughtering operations has been identified as a potential source of human pathogens into the animal food chain (Bell, 1997; McEvoy et al., 2003; Pearce et al., 2004; Food and Agriculture Organization (FAO), 2009; Hue et al., 2010; Bello et al., 2011; Zweifel et al., 2015; Hazards (BIOHAZ) et al., 2021; Klaharn et al., 2022), but the resultant or associated food safety risks have so far, been poorly characterized. Studies have speculated or identified the most tangible or probable source(s) of pathogenic contamination to meat along the slaughtering continuum, but many collected samples for microbiological analyses and/or clonal tracing from slaughterhouse paraphernalia, equipment, surfaces,

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input, even drinking water, but not process water (Lawrence & Gilmour, 1995; Klein et al., 2007; Magistrali et al., 2008; Damjanova et al., 2011; Melero et al., 2012; Sala et al., 2016; Marin et al., 2022). There is consequently a dearth of constructive studies describing the microbiological hazards in abattoir process-water, as well as the associated food safety risks.

Interventions to meaningfully minimize meat consumers' exposure to pathogens through water used for abattoir operations depend on correctly understanding its role as a significant contributor to pathogen contamination in the animal production system (Doyle & Erickson, 2012). This overview discusses the relevance of slaughterhouses and water used in slaughtering operations for the microbiological safety of meat. It explains the need to characterize food safety risks associated with pathogens introduced to carcass through water at the slaughter stage and then proposes a framework for the design of studies to establish water as a vehicle of pathogens to carcasses at the slaughterhouse stage. Finally, it offers some research recommendations and technical guidance for the characterization of associated food safety risks.

Slaughterhouses as a potential source of foodborne pathogens for food animal

Many important meat-associated pathogens, including *Campylobacter* introduced to livestock on the farm, may become established and proliferate in their gut (Borch et al., 1996; Hermans et al., 2011; Aditya et al., 2023). Control efforts have thus largely prioritized mitigation at the primary production stage. Studies show that preharvest interventions effectively reduce pathogen prevalence in herds and flocks (Newell & Fearnley, 2003), and such approaches continue to be valuable components of contemporary, comprehensive food safety strategies. However, preharvest control represents only one point along the production continuum where contamination can occur. Even with effective on-farm management, subsequent processing stages, such as slaughterhouses, are potential opportunities for pathogen introduction or cross-contamination that warrant research attention.

Studies have highlighted the importance of environmental sources of pathogens in animal food chains (Lawrence & Gilmour, 1995; Rugna et al., 2021). In particular, research has underscored the role of the slaughterhouse environment in microbial contamination of meat (Peruzy et al. 2021; Rugna et al., 2021; da Silva & Farag, 2021). Sakaridis et al. (2011) reported 60% *Listeria monocytogenes* prevalence in poultry samples from a slaughterhouse with *L. monocytogenes*-contaminated surfaces, compared to 16–40% pathogen prevalence in facilities without environmental contamination. Notably, some studies have recovered pathogens from slaughterhouse environments even when they were not detected in farm animals or farm inputs. For example, Sereno et al. (2019) did not detect *L. monocytogenes* in farm animals, but *L. monocytogenes* was recovered from slaughterhouse samples. Morales-Partera et al. (2018) detected indistinguishable Pulsed-Field Gel Electrophoresis (PFGE) patterns of *L. monocytogenes* in pigs sourced from different farms but slaughtered in the same abattoir on separate days—strongly implicating the slaughterhouse itself, rather than the animals, as the contamination source. These findings indicate that slaughterhouses can serve as independent sources of contamination regardless of the microbiological status of incoming animals.

Studies have also shown that slaughterhouses are a critical control point in the meat production chain, where even low levels of microbial contamination can be amplified and disseminated downstream, ultimately compromising the safety of finished products (Belluco et al., 2016; da Silva & Farag, 2021; Rossler et al., 2020; Nastasijevic et al., 2023). A growing body of evidence demonstrates that foodborne pathogens such as *L. monocytogenes*, *Salmonella*, and *Campylobacter* can persist in slaughterhouse environments, and potentially establish long-term reservoirs that contribute to persistent contamination (Lawrence & Gilmour, 1995; Lucchini et al., 2023). For example, Lawrence and Gilmour (1995) used random amplification of polymorphic DNA (RAPD) and multilocus enzyme electrophoresis to characterize *L. monocytogenes* isolates from poultry products and processing environments, and demonstrated that the same strains persisted in the processing environment and repeatedly contaminated products.

Tracking studies have confirmed that certain pathogen strains endure or persist in spite of cleaning and disinfection protocols, enabling them to colonize equipment, drains, and surfaces for weeks or months (Sereno et al., 2019; Marin et al., 2022). Zeng et al. (2021) found that *Salmonella* was still detectable on poultry-slaughter equipment after cleaning and disinfection, with positives concentrated in early-process areas such as hanging and scalding. Contaminated equipment and transport crates were shown to transfer *Salmonella* to carcasses from officially negative flocks, including persistent serovars such as *S.* Infantis and *S.* Paratyphi B var. Java. Similarly, Hernández et al. (2013) reported that different *Salmonella* serotypes were isolated at various stages along the slaughter line, including in trucks and lairage, even after cleaning and disinfection measures were implemented. These findings underscore that slaughterhouses are not merely passive conduits but active determinants of the microbiological safety of meat.

Interventions targeting slaughterhouse hygiene have demonstrably reduced public health risks, reinforcing the argument that when slaughterhouses are properly recognized and managed as critical control points, meaningful improvements in food safety can be achieved. For example, stricter requirements for the presentation of clean cattle at slaughter are hypothesized to have contributed to reducing the risk of foodborne transmission from beef, although contemporaneous field, slaughterhouse, and end product data have not demonstrated this directly or significantly. (Bishop et al., 2022).

Water as a potential reservoir and vehicle for foodborne pathogens to carcass in slaughterhouses

The microbiological status of livestock entering slaughter can vary considerably among batches depending on herd status, transportation conditions, and lairage practices (Arguello et al., 2013). Once animals enter the slaughter process, microbial contamination may arise from the animal itself, personnel, the environment, persistently contaminated surfaces and equipment, or water (Nógrády et al., 2008; Choi et al., 2013; Hernández et al., 2013; Ferreira et al., 2014; Rouger et al., 2017).

Although meat processing varies by animal type, for instance, poultry carcasses typically undergo immersion-based washing while beef and pork carcasses are spray-washed, direct contact with water is common regardless of species. Water is integral to processing but also to most decontamination protocols, including washing with organic acid solutions, sanitizers, chlorinated water, or hot water, as well as sterilizing utensils (Bell, 1997; Cabral et al., 2017; da Silva & Farag, 2021; Wambui et al., 2018).

Like other environmental niches, water can harbor a wide variety of human pathogens (Ellerbroek et al., 2010; Klaharn et al., 2022), making microbiological hazards in process water a potential contamination risk for carcasses.

Water-mediated contamination and variable decontamination outcomes

Research evidence indicates that water can act as a decontamination medium but also, under the right conditions, as a vehicle for pathogen transfer to carcasses. Studies report measurable reductions in indicator organisms following washing or hot water/steam treatments, with reported reductions up to ~1 log for *E. coli* and aerobic bacteria, and higher reductions when hot water or steam pasteuriza-

tion is applied (Castillo et al., 1998a; Bosilevac et al., 2006; Belluco et al., 2016).

Multiple studies however also suggest that water may either introduce pathogens onto carcass surfaces or facilitate cross-contamination and redistribution of microorganisms on carcass surfaces, including transfer to previously uncontaminated areas (Castillo et al., 1999; da Silva & Farag, 2021). Bello et al. (2011) observed significant postwash increases in *E. coli* levels ($P < 0.05$), from log 4.3–4.6 cfu/cm^2 to log 4.6–4.9 cfu/cm^2, and suggested a role for water quality in contamination outcomes. McEvoy et al. (2003) found that carcass parts initially negative for *E. coli* O157:H7 became positive after washing, consistent with washwater-mediated contamination reported by others (Bell 1997; Castillo et al., 1998b; Júnior et al., 2024). Some other studies have also shown that subsequent water-based steps can recontaminate carcasses even after prior decontamination (Yu et al., 1999; Zweifel et al., 2015; Althaus et al., 2017), with some findings suggesting that risks may be related to specific operations along the slaughter line (Althaus et al., 2017). Bell (1997) found that cold water washing did not effectively remove microbial contamination but redistributed it, increasing microbial counts in some areas. Conclusions of some studies, in fact, indicated that washing has little microbiological benefit despite improving the physical appearance of carcass (Gill et al., 1996; da Silva & Farag, 2021).

Overall, the effectiveness of water-based interventions—and the potential for microbial contamination or unintended redistribution of contamination—appears highly context-dependent. Outcomes vary depending on microbiological quality of water, temperature control, disinfectant type and concentration, application point along the slaughter line, spray pressure, and opportunities for or the ability of bacteria to persist in protected microniches. Process design may also influence the magnitude of risks for microbial transfer. In poultry systems, where immersion-based scalding and washing steps are common, poor water quality or inadequate temperature control may facilitate widespread redistribution of microorganisms. In contrast, spray washing of red meat carcasses may localize transfer but still permit pathogen spread when water quality, pressure, or reuse are suboptimal. These process-specific differences may explain why washing can, under certain conditions, increase rather than reduce microbial contamination.

Hot water applications and limitations

Hot water has been experimentally validated to reduce microbial levels on chicken, lamb, and sheep carcasses (Hugas & Tsigarida, 2008; Tompkins et al., 2008; Loretz et al., 2010; Zhang et al., 2013). Kelly et al.(1981) achieved significant reductions (>1 log$_{10}$/cm^2) in aerobic plate counts on lamb carcasses sprayed with water at temperatures ≥80 °C, while Gorman et al. (1995) demonstrated that increasing spray wash temperature from 16 to 35 °C to 74 °C more efficiently removed *E. coli* and visible fecal contamination from beef tissue. Also, Klaharn et al. (2022) reported statistical relationships between poor scalding temperature control and noncompliant indicator counts on chicken carcasses.

Despite these benefits, hot water or steam interventions are not always feasible in commercial settings because they may compromise product quality. Laboratory studies have reported heat-associated damage to carcass surface tissues (Bolton et al., 2001; Loretz et al., 2010). These constraints can limit the adoption of hot water or steam-based interventions in some slaughterhouses, despite their potential for superior microbial reduction.

What is the risk burden? Does the extent of risks warrant regulatory enforcement for water used for slaughtering operations?

In many developed countries, such as the United States, water used in meat processing that has direct or indirect contact with meat is expected to be of drinking standard (National Advisory Committee on Microbiological Criteria for Foods, 2022); thus, regulations in those areas require and largely enforce or at least recommend the use of potable water for slaughtering operations. In some other countries, such as in New Zealand, processors commonly apply antimicrobial intervention steps such as high scald temperatures (Kingsbury et al., 2023). In Australia, meat processing plants apply hot water treatment, while in the EU, only potable water that is hot or steam pasteurized, as well as lactic acid carcass washing, is permitted for use in abattoirs (Antic et al., 2021). Water quality guidelines designed by Chinese authorities specify that the water used for carcass processing shall meet the national drinking water standards or relevant standards for water quality. The water supply system shall be designed, constructed, and maintained to prevent contamination and ensure the safety and hygiene of the water used in processing. The standard also includes guidelines for the drainage of wastewater and the centralized processing of industrial wastewater to prevent environmental pollution (National Standard of the People's Republic of China, 2016). Additionally, the WHO stipulates that water from alternative sources that would come in indirect or direct contact with the product must meet drinking water standards (Bello et al., 2011). Although many developed countries enforce the use of potable water in slaughterhouses, global standards for water quality and pathogen control during meat processing are inconsistent. This gap raises concerns about the adequacy of existing regulations in mitigating relevant risks.

To satisfy international meat trade and export standards, available data indicate that many producers tend to adhere to these regulations and guidelines (Codex Alimentarius Commission, 2005; Mabunda et al., 2025). In spite of these guidelines and regulations in many parts of the advanced world, enforcement for water used in slaughterhouses is much less stringent compared to that for water used in some other food production chains, such as cultivation of fresh plant produce. For example, in China, enterprises are expected to control water quality (National Standard of the People's Republic of China, 2016). This disparity is likely due in large part to the fact that irrigation water has been firmly established as a potential source of human pathogens to produce on-field (Steele & Odumeru, 2004). It could also be due to the fact that meat is in many parts of the world, typically subjected to a cooking step prior to consumption, which provides an additional lethality barrier that may influence risk perception and regulatory emphasis/consideration compared with foods commonly consumed raw.

In addition, due to many related foodborne outbreaks and industry recalls, associated food safety risks have been extensively studied (Uyttendaele et al., 2015; Rock et al., 2019; Alegbeleye & Sant'Ana, 2023). Research has characterized and described the behavior of human pathogens introduced to produce via irrigation water, including the potential and associated dynamics for attachment and or internalization into produce commodities (Warriner et al., 2003; Ibekwe et al., 2004; Oliveira et al., 2011). Other factors that modulate the risks for the introduced pathogens to constitute food safety hazards, such as the potential for survival and or proliferation in produce commodities and products (e.g. Fresh Cut Vegetable salads) have also been studied (Van der Linden et al., 2013). Consequently, the need to use microbiologically safe water for growing vegetables is widely acknowledged, advocated, and even enforced in some parts of the world, with the US FDA FSMA rule on produce safety (US FDA, 2020c) as a good case in point.

In spite of acknowledged risk potential, the microbiological safety requirement for water used for slaughtering operations to minimize food safety risks is not yet clear. For instance, is it necessary that regulatory authorities enforce the use of potable water for washing and other relevant processes in slaughterhouses? Are global standards, benchmarks, or thresholds for selected microbiological parameters required? Or should the industry continue to rely on a combination of general food safety principles (like HACCP), national regulations,

and nonbinding international guidelines to manage water quality on a site-specific, risk-assessment basis? Should the use of hot water be mandatory, wherever possible? To constructively address these safety questions, empirical data demonstrating the food safety risks associated with the use of microbiologically unsafe water in slaughterhouses are required. In the next section 5, therefore, we discuss first, the need to characterize food safety risks associated with the application of microbiologically unsafe water in slaughterhouses, and then in subsequent sections 6 and 7, we offer some suggestions for an appropriate study design.

Why is it necessary to measure food safety risks associated with enteric bacteria introduced to meat via water during the slaughter process?

Although there is no research evidence suggesting that water may introduce peculiar or potentially more severe hazards to meat in slaughterhouses, there is also no verifiable evidence that associated hazards can be taken as benign. For one, the type(s) of microbiological hazards that may be introduced to the carcass especially but also the meat processing environment from the slaughtering point through water, have not been properly characterized. Some strains of pathogens, such as *Bacillus cereus* and *Staphylococcus aureus,* which have been previously associated with water, produce virulent, heat − stable enterotoxins that can persist and retain virulence in spite of heat treatment (Abdou et al., 2012; Trigui et al., 2015). Some of these pathogens have exhibited remarkable potential to survive in water (Thomas et al., 1999; Cools et al., 2003) and have been detected along animal food chains. What is, however, not clear is whether or not water used for slaughtering processes is a peculiar source of such pathogens along animal food chains in various parts of the world. It might be useful to clarify here that, while water can potentially introduce pathogens such as *Bacillus cereus*, including enterotoxin − producing strains into the meat chain, there are several other potential sources of enterotoxin − producing *Bacillus* spp. along the meat production–distribution continuum. Additionally, it should be emphasized that there is, thus far, no rigorous surveillance or scientific evidence indicating that water is a peculiar route of these types of pathogens to carcass, but there is also no scientifically valid reason to doubt that water may, via this form, contribute to food safety risks.

Since the behavior of pathogens originating from water has so far not been robustly characterized, it is also not yet clear if pathogens originating from water may persist more adroitly along the production-processing continuum and pose specific or more significant risks for foodborne illnesses. Already, there is some research indicating that microbial isolates of environmental origin have superior capacity to adapt; for example, form or participate in biofilms, resist antimicrobials, survive established cleaning protocols, or otherwise persist in processing plants or along the food chain (Lawrence & Gilmour, 1995; Šimunović et al., 2020). For instance, Caruso et al. (2020) reported that the majority of *L. monocytogenes* isolates (serotypes 4b/4e and ST2) recovered from dairy, fish, and meat production chains that exhibited elevated MIC (Minimum Inhibitory Concentration) to oxacillin were of environmental origin. Takahashi et al. (2006) and Klein et al. (2007) found that of a highly diverse set of isolates recovered along the production/processing line and slaughterhouse, respectively. *C. jejuni* genotypes that were most tolerant to environmental stress were able to survive the slaughtering process and contaminate final meat cuts. Growth conditions encountered by bacteria prior to entering a new environment, such as the animal food chain, may influence whether or not the cells will become successfully established in the new niche. Previous growth history plays a major role in the preadaptation of bacterial cells (Rozen & Belkin, 2001) and pathogens can evolve genetic mechanisms that influence their fitness in environmental and food matrices, partly due to their prior growth conditions. For instance, some cold − tolerant microorganisms may have evolved in part due to prolonged exposure to cold temperature conditions (Hingston et al., 2019).

There are specific examples of studies in the literature that found that the source of the isolate influenced the growth kinetics/behavior. For example, Begot et al. (1997) found that compared to isolates of *L. monocytogenes* from meat products, isolates from industrial sites were faster growing in tryptic meat broth. Similarly, Hudson (1992) found that food-derived (from ready-to-eat fleshfood sources) strains of *Aeromonas hydrophila* were better adapted to growth at lower temperatures, compared to those obtained from meat processing or clinical sources. These findings indicate that it is useful to assess the growth behavior of water-sourced pathogens along the meat chain. Admittedly, research data on behavior of foodborne pathogens introduced into the meat chain from all possible sources, not only water, are desirable (Choi et al., 2013), but also the behavior of water − associated pathogens in animal food chains should not be overlooked. For context and emphasis, due to climate change, access to safe, inexpensive water is becoming increasingly fraught and, based on predictions and estimates, may be under significant threat. Food animal processors may, due to shortages, turn to alternative (probably suboptimal quality) water sources, potentially increasing associated food safety risks.

Food safety risks from waterborne pathogens in slaughterhouses

Pathogens introduced to meat through slaughterhouse water can pose varying degrees of food safety risks depending on several factors, including their ability to persist, be inactivated by bactericidal treatments, or survive until human consumption. Pathogens may become inactivated, reduce in concentration, or persist, leading to potential contamination of meat products consumed by humans. The framework provided in Figure 1 elaborates some steps or procedures that can help relevant experts to unequivocally or at least meaningfully determine food safety risks associated with water used for slaughterhouse operations. Relevant associated details for elucidated steps follow in section 7 – 7.3 below. A relevant, related risk that must remain in perspective is the potential fate of human pathogens introduced via other routes or sources, that abattoir wash-water or water-related procedure fails to remove from the carcass. It seems likely that the potential range of possible eventualities (Table 1) applies to these pathogens.

Research priorities for characterizing waterborne pathogen risks in slaughterhouses

Implementing effective control measures to mitigate or manage the issues identified in sections 2 – 6 above requires focused research in three priority areas (Table 2). These include characterizing water quality in slaughtering operations, understanding the dynamics of pathogen transfer from water to carcass, and tracing the fate of waterborne pathogens through the processing and distribution chain.

Characterizing water quality in slaughtering operations

Effective management of food safety risks requires comprehensive microbiological assessments of water used in slaughterhouses. To better inform control measures, more studies should identify pathogen profiles and concentrations in process water. While high temperatures in scald tanks typically reduce the incidence of pathogens, occasional exceptions exist, which highlights the need for continuous monitoring (Gruntar et al., 2015; Hauge et al., 2023).

Pathogen transfer from water to carcass

Although process water contamination does not always result in carcass contamination, transfer mechanisms exist where pathogens

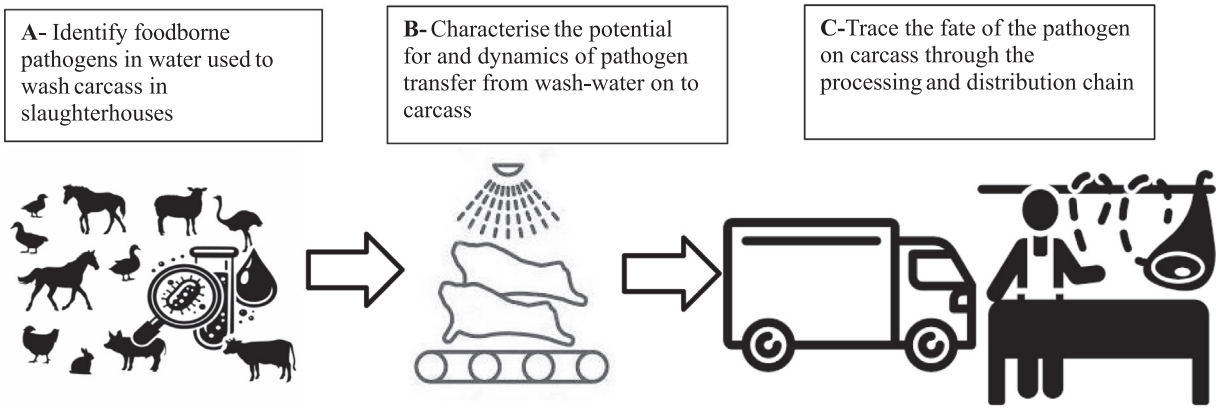

Figure 1. A framework for characterizing foodborne illness risks associated with the application of microbiologically contaminated abattoir wash water.

Table 1
Potential fate of human pathogens on carcass

Bacteria on carcass may occur as:	What may happen (fate)	Lower-risk conditions (more likely inactivation/reduction)	Higher-risk conditions (more likely persistence/growth/spread)
Planktonic cells	May be reduced, redistributed, persist, or spread via water movement	Clean water, controlled application (time/temperature), effective sanitation	Poor water quality, reuse/recirculation, organic load → redistribution/cross-contamination
Surface-attached cells	May persist through processing and remain detectable after washing	Adequate mechanical removal + validated interventions	Incomplete coverage, shielding in micro-niches, high initial load
Biofilm-associated cells	Can seed recurring contamination and recontaminate carcasses	Effective sanitation of high-contact points, hygienic design	Persistent niches (drains, joints), inadequate sanitation → recurrent contamination
Cells in protected sites (e.g., tissue folds, cut surfaces, beneath fascia/fat)	More likely to persist into downstream handling	Proper hygienic dressing, preventing cross – contamination early	Early contamination + protection by tissue/fat → persistence during storage/ processing

Table 2
Proposed framework to characterize food safety risks associated with waterborne pathogens in slaughterhouses

Step	Objective	Activities	Expected outcome
Step 1: Characterization of water quality	Identify and quantify microbiological hazards in water used for processing carcasses.	Sample collection and analysis of water from slaughterhouses. Measure pathogen profile and concentration. Assess physicochemical properties of water.	Data to guide water management practices and control measures in slaughterhouses. Identification of high-risk pathogens in water sources.
Step 2: Evaluate (risks for) pathogen transfer to carcass	Assess how pathogens in water transfer to carcasses and the dynamics of contamination.	Monitor pathogen concentration on carcasses postwash. Analyze genotypic relationships between waterborne pathogens and those found on carcasses.	Clear understanding of how water contamination leads to the incidence of pathogens on carcasses. Identification of critical points for controlling contamination
Step 3: Trace pathogens through the processing chain	Monitor pathogen persistence throughout processing and distribution until retail.	Trace pathogen contamination from the slaughterhouse to the final meat products. Conduct molecular tracking of pathogens through the supply chain.	Insights into how pathogens survive through different processing stages. Better risk assessments for foodborne illnesses due to persistent pathogens in the supply chain

in water reach meat surfaces. Experiments that establish genotypic relationships between pathogens in water and those found on carcasses can confirm the role of water as a vehicle for pathogen transfer (Althaus et al., 2017; Stopforth et al., 2006).

Tracing the fate of pathogens through the processing and distribution chain

Once pathogen transfer from water to carcass has been established (Section 7.2, Step 2), the next critical research priority — corresponding to Step 3 of the proposed framework (Table 2) — is to trace the fate of these pathogens through subsequent processing and distribution stages until retail. This involves monitoring pathogen persistence at each stage and conducting molecular tracking (e.g., using PFGE, Multilocus Sequence Typing [MLST], or whole-genome sequencing) to determine whether waterborne isolates recovered at the slaughterhouse can be detected on final meat products. Such studies would provide insights into how pathogens survive different processing stages and would support more robust risk assessments for foodborne illnesses attributable to persistent waterborne pathogens in the supply chain (Melero et al., 2012). The subsections below discuss two important factors that influence pathogen fate along the processing chain: persistence potential (Section 7.3.1) and antimicrobial resistance (Section 7.3.2).

Pathogen persistence in slaughterhouses. Persistence of foodborne pathogens in slaughterhouse environments is influenced by their ability to adapt to stressors such as heat and antimicrobial treatments (Yousef & Juneja, 2002). Some pathogens, like *L. monocytogenes* and *Campylobacter*, have exhibited resistance to common inactivation

strategies. The persistence potential depends on factors such as intrinsic pathogen traits and environmental conditions (Marmion et al., 2022; Lucchini et al., 2023).

Antimicrobial resistance. Antimicrobial-resistant pathogens pose additional risks. Resistant strains of *Listeria*, *Salmonella*, and *E. coli* have been found in slaughterhouses and along the meat supply chain (Nastasijevic et al., 2024). There is increasing concern over resistant pathogens entering the chain through contaminated water sources (Sereno et al., 2019; Dantas et al., 2020; dos Santos et al., 2022).

Other research needs/considerations

While laboratory – scale studies provide meaningful output, studies should be conducted in commercial environments with naturally contaminated carcasses. Porcine, cattle, sheep, goats, and poultry slaughterhouses are some of the most commonly studied in the literature. All types of slaughtering activities, including those that kill rabbits for food as well as polyvalent slaughtering activities, deserve research attention. It might also be interesting to attempt to establish whether slaughterhouses, as pertaining to the microbiological safety of water used, represent a more vulnerable point compared to other points along the meat chain. Other research areas to explore include assessing whether abattoirs and abattoir operatives appreciate the risks associated with the use of marginal or more specifically, microbiologically contaminated water. Based on the review of the literature, there needs to be more surveys to understand the attitude of abattoir workers and other relevant stakeholders along the continuum, including vendors. If pathogens introduced to the carcass through water are characterized to be able to persist up till the point of consumption, it might be necessary that the water used at this point, for these purposes are subject to the same or similar regulatory stipulations as irrigation water used to cultivate vegetables. Strain variabilities in the capacity to persist have been widely described, and we suggest that experiments determining the response of bacteria to test inactivation or reduction technologies take cognizance of this.

In the overview by Fegan et al. (2022), a list of parasites, including protozoan parasites, matched with the corresponding typical meat vehicle, is presented. Waterborne parasites have also been isolated from various types of water resources, including those that could be potentially exploited for slaughtering operations, and this means that this area of research deserves some more attention. Studies should thus screen for parasites in abattoir washwater provisions, but also determine the behavior of viruses and parasites introduced to carcass and slaughterhouse environment via water.

Conclusions

This paper reviews the available evidence on the potential for water used for slaughtering operations to transfer microbiological hazards to carcass. Studies reviewed show that water can introduce pathogens to meat during slaughtering and may also serve as a medium for the dissemination of pathogens on various parts of carcass. Water used in slaughterhouses can influence the microbiological safety of food animals, but the relevance of this route of contamination, as well as the food safety risks associated with contamination, needs to be rigorously quantified. The fate of foodborne pathogens introduced to carcass at slaughterhouses through water significantly influences the risks of transmission to humans. The behavior of foodborne pathogens, with particular focus on the potential and mechanisms of bacterial persistence, was thus explored. The need for further research on the persistence of foodborne pathogens in meat and meat products, as well as in the slaughterhouse environment, was identified. Consumer behavior, such as poor handling or inappropriate cooking practices, can amplify risks. Characterizing the magnitude of relevant food safety risks requires, in part, measuring or gauging the susceptibility of introduced microbial agents to common inactivation techniques such as cooking. It is, therefore, vital to assess consumer behavior on all relevant aspects. In addition to experimental and technical approaches, therefore, optimized methodologies to properly assess human behavior contextually are required. All processing steps and input, along with slaughtering, are important, but water as a potential food safety hazard in slaughterhouses should not be overlooked.

Ethics approval and consent to participate

This manuscript has undergone the required approval that assures the research complies with ethical standards and maintains the integrity of the research. As a review article, the rights and welfare of participants are not applicable.

Statements and declarations

Nothing to disclose.

Consent for publication

Granted as required.

Availability of data and material

Not applicable.

CRediT authorship contribution statement

Oluwadara Alegbeleye: Writing – review & editing, Writing – original draft, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Javier Mateo-Sagasta:** Writing – review & editing, Visualization, Validation, Supervision, Resources, Project administration, Methodology, Funding acquisition, Conceptualization.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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