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Listeria Occurrence and Potential Control Strategies in Alternative and Conventional Poultry Processing and Retail

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Listeria monocytogenes is a psychrotrophic Gram positive organism that is considered one of the more critical foodborne pathogens of public health concern. To prevent illness the USDA and FDA enforce a zero-tolerance policy for *Listeria* on ready-to-eat foods such as delicatessen meats and poultry. Regardless, *L. monocytogenes* can still be isolated from food production facilities and retail products, indicating that current sanitation methods are not always sufficient. Both conventional and alternative poultry production and processing systems have also been identified as potential sources of *Listeria* spp. Concerns associated with alternative poultry production and processing can be further exacerbated by limitations on sanitation and available antimicrobials for usage in organic and natural poultry products. Furthermore, mobile poultry processing units often process organic and small-scale poultry farms that are not able to be processed by conventional standing facilities. These alternative production facilities and their products are often exempt from federal inspection, due to processing a relatively low number of carcasses. Due to these exemptions, it is unknown if sufficient sanitation is applied in these alternative processing facilities to prevent *L. monocytogenes* contamination. Organic processing restrictions may also impact which sanitizers and antimicrobials can be utilized. This review describes variations between conventional and mobile poultry processing units in conjunction with how *L. monocytogenes* may persist in the processing environment and on retail products. This review will also examine alternative antimicrobials proven to be effective against *Listeria* spp. and potentially be acceptable for use in alternative poultry production systems.

Keywords: retail products, mobile poultry processing, alternative poultry production systems, conventional poultry, poultry, *Listeria*

INTRODUCTION

Listeria monocytogenes is a Gram-positive, facultative anaerobe that is the causative agent of listeriosis (Gudbjörnsdóttir et al., 2004). They are considered saprophytic organisms with the capability to adapt to an ever-changing environment because they possess multiple stress response mechanisms to overcome varying temperatures, salt concentrations and pH (Berrang et al., 2000; Milillo et al., 2012a; Giaouris et al., 2015; Saldivar et al., 2018). *Listeria monocytogenes* infection is

especially concerning to the elderly, immunocompromised, and pregnant women who are most susceptible and mortality rates can exceed 20% (Ryser and Marth, 2007; Tsai et al., 2011; Milillo et al., 2012a; Silk et al., 2013; Centers for Disease Control Prevention., 2016).

Due to variations in ecology, genomic content, and recombination rates four distinct evolutionarily lineages have been described for *L. monocytogenes* (Zhu et al., 2005; Milillo et al., 2012a). Lineage I contains the serotypes 1/2b, 3b, 3c, and 4b, and are differentiated based on cell wall expression (Gray et al., 2004; Nightingale et al., 2005; Zhu et al., 2005). Lineage II is composed of serotypes 1/2a, 1/2c, and 3a (Orsi et al., 2011). Lineage I is responsible for the majority of human outbreaks, but a majority of food isolates of *L. monocytogenes* are from lineage II (Nadon et al., 2001; Nightingale et al., 2005; Orsi et al., 2011; Tsai et al., 2011). Lineage III and IV are not typically relevant clinically for humans (Orsi et al., 2011).

With listeriosis estimated to cause 23,000 illnesses annually, it is imperative that intervention strategies be established to reduce the risk of foodborne illness to consumers, specifically *L. monocytogenes* (Noordhout et al., 2014). When factoring in demographic changes in the U.S., such as age and race, the rate of listeriosis is expected to increase from 0.25 per 100,00 in 2010 to 0.32 in 2030 (Pohl et al., 2017). *Listeria monocytogenes* can proliferate and survive within the food processing environment, and ready-to-eat (RTE) meats becoming a potential source of illness when the food product is consumed directly without further preparation (Lavieri et al., 2014; Crandall et al., 2015). Previous research has shown contamination by *L. monocytogenes* of RTE foods mainly occurs during slicing (Ryser and Marth, 2007) and packaging after cooking because these products are usually cooked during processing and consumed without further cooking [U.S. Department of Agriculture Food Safety Inspection Service (USDA-FSIS), 2006]. Contamination of these types of foods can lead to major outbreaks of listeriosis. For example, in 2008 an outbreak of listeriosis in Canada linked to delicatessen meats caused $242 million (CSD) dollars in damage and 24 deaths (Thomas et al., 2015). These products include poultry deli meats. *Listeria* spp. plate counts are considered a good indicator of potential *L. monocytogenes* contamination due to similar growth characteristics (Food Safety Inspection Service, 2012). Concern over potential foodborne infection has led to a zero-tolerance policy on *Listeria* spp. in RTE products by the United States Department of Agriculture (USDA) and the Food and Drug Administration (FDA) (Shank et al., 1996).

Additionally, *L. monocytogenes* can also be a concern for poultry contamination (Silk et al., 2013). Despite the stringent policies, *L. monocytogenes* can be isolated from poultry products and 1,651 cases of listeriosis and 346 deaths were reported from 2009 to 2011 (Silk et al., 2013). Grow-out farm studies have indicated that broilers can be an important vector for *Listeria* spp. contamination of the processing environment (Iida et al., 1991; Rothrock et al., 2017). A study of a poultry processing plant indicated that the raw product was a potentially important source of *L. monocytogenes* (Berrang et al., 2010). However, corresponding studies to the best of the authors' knowledge have not been performed for alternative poultry processing even

though *Listeria* spp. can be isolated from pasture-raised poultry (Milillo et al., 2012b; Locatelli et al., 2017).

Other factors may play a role in increasing listeriosis risk depending on the poultry processing management system. For example, organic poultry production is not permitted to apply commonly used synthetic antimicrobial agents including nitrates and nitrites, which can inhibit *L. monocytogenes* (Winter and Davis, 2006; Sullivan et al., 2012; McDonnell et al., 2013). As this sector of the poultry industry grows, the limited choices of antimicrobial agents could become problematic and lead a greater exposure risk associated with these types of products. This risk could increase as this market sector continues to grow in popularity. In 2016, the organic poultry product market was $750 million and trends indicate a sharp increase in market share (Philips, 2017). This sharp increase was corroborated by the 2017 market share of organic poultry reaching $1 billion and 2018 sales expected to reach 1.2 billion (Organic Trade Association., 2018). In 2017, the total organic market represented over $50 billion in sales (Organic Trade Association., 2018).

In addition, the pasture-raised or free-range poultry industry continues to expand into the market place and in turn, presents a new set of challenges for food safety (Van Loo et al., 2012). Some of these flocks may be processed using a mobile poultry processing unit (MPPU) (Trimble et al., 2013; O'Bryan et al., 2014; Micciche et al., 2018a). Other alternative small scale processing facilities are available but due to the paucity of pathogen control information will not be discussed further (Fanatico, 2003; Ahuja and Sen, 2007; Trimble et al., 2013). In the current review, after a brief description of non-conventional poultry processing systems, the focus will shift to the opportunities and documented occurrence of *Listeria* spp. in conventional and alternative poultry processing systems. Emphasis will also be placed on the options for controlling pathogenic *Listeria* in alternative poultry processing operations where intervention choices are more limited.

POULTRY PROCESSING SYSTEMS AND ENVIRONMENTS

In 2017, over 41.6 billion pounds of chicken was produced in the U.S. and this is possible through a streamlined processing system operating at 140–175 birds/min (9 CFR 381.69) [Owens et al., 2000; United States Department of Agriculture (USDA), 2018]. The conventional poultry processing system has been extensively described in reviews by Owens et al. (2000), Sofos et al. (2013), and Blevins et al. (2018) and will only briefly be covered here. Birds that have been feed withdrawn are brought to the processing facility where they are hung on an automated line, stunned, and exsanguinated (Smith, 2014; Barbut, 2016). The carcasses are subsequently heated in a scalder (56–65°C) and are de-feathered in a picker (Sofos et al., 2013). The scalder uses ∼1 L of water per bird (Barbut, 2016) and has been noted as a potential source of cross-contamination and investigated in Rothrock et al. (2016a). After a chlorine or peracetic acid (PAA) wash, undesired body parts and internal organs are removed (Sukted et al., 2017; Blevins et al., 2018). The careful removal of the gastrointestinal

tract (GIT) during evisceration is important as rupturing of the organs, such as the ceca or crop, can introduce bacteria and potential pathogens to the product. Following evisceration, birds are washed with an inside-outside bird washer and are typically placed in a pre-chiller for 10–15 min followed by immersion in the main chiller for 45–115 min at 4°C (Sams and McKee, 2010; Blevins et al., 2018). Further processing and packaging may occur which depends on the desired product (Sofos et al., 2013). However, alternatives exist to these centralized facilities, such as MPPU (Micciche et al., 2018a).

Mobile poultry processing systems are employed to supplement the limitation of centralized processing facilities (Ollinger et al., 2005; Van Loo et al., 2013; O'Bryan et al., 2014). With a 23% decrease in the number of slaughterhouses from 1992 to 2008, there is an increased geographical restriction from some farms and processing centers that make conventional processing impractical (Zezima, 2010; O'Bryan et al., 2014). Furthermore, due to limitations on the number of birds processed without the need for inspection small scale MPPUs are also desirable (New Entry Sustainable Farming Project, 2012; Van Loo et al., 2013). Mobile poultry processing units have been extensively reviewed by O'Bryan et al. (2014) and Micciche et al. (2018a) and only a limited discussion will be provided in the current review. Mobile poultry processing units are typically 5–11 m long and can be transported on trailers. Trailers with the MPPUs have a similar processing step flow to conventional poultry processing systems (O'Bryan et al., 2014). However, there are practical differences that may impact how pathogens are remediated.

Design choice is highly variable in MPPUs (New Entry Sustainable Farming Project, 2012). While some may employ a shackle hang system for the carcasses, many utilize kill cones with blood troughs for exsanguination (Fanatico, 2003). If the cones are not properly cleaned, cross-contamination events may occur early in the processing. From the kill cones or shackle hang system, carcasses are transferred to a scalder, which may be as simplistic as a bucket of hot water, and then plucked and placed in a holding container, usually made of plastic (New Entry Sustainable Farming Project, 2012; O'Bryan et al., 2014). In conventional systems, these processes are typically separated from downstream processing by a physical barrier due to the blood and offal waste produced (Keener et al., 2004; Angioloni et al., 2016). This barrier may not be present in MPPU's, although Mancinelli et al. (2018) describes an Italian MPPU with a barrier in place.

Once available, workers eviscerate the carcass and remove inedible components including the GIT on evisceration tables (New Entry Sustainable Farming Project, 2012). Like the kill cones, cross-contamination events can occur here if the table is not appropriately cleaned between eviscerations. After evisceration and subsequent washing, carcasses are placed in a chill tank which can be simply an ice bucket. After the carcass is chilled, birds are further processed and deboned which in some MPPUs is done on a polyvinyl chloride (PVC) framework (New Entry Sustainable Farming Project, 2012).

Another variation in design includes whether an MPPU is open or enclosed (New Entry Sustainable Farming Project, 2012). Enclosed facilities may suffer from the need to appropriately clean the facility in the case of splashing, while open-aired facilities have to contend with inclement weather (O'Bryan et al., 2014). From an environmental standpoint, there are concerns regarding contaminated processing water overflowing onto the ground around the open-aired MPPU (New Entry Sustainable Farming Project, 2012). Additionally, despite state regulations, wastewaters from MPPUs have been applied to gardens as fertilizer or discharged onto private property (Fanatico, 2003; Hoppe, 2010; O'Bryan et al., 2014). This may be a concern if pathogens, such as *L. monocytogenes*, are present in the processing water and if dumped near the farm it has the potential to be reintroduced into successive flocks (O'Bryan et al., 2014; Micciche et al., 2018a).

While non-conventional poultry processing differs from conventional poultry processing, opportunities still exist for *L. monocytogenes* contamination. In MPPUs floor drains are not always present and, in closed trailer environments, there is a potential for worker exposure that may be transferred to downstream processing provided the worker operates at multiple stations (Mancinelli et al., 2018). Organic poultry processing may have additional restrictions that limit the use of antimicrobials and may, therefore, restrict the use of sanitizers in MPPUs. Lactates and diacetates were prohibited in both the processing and final packaging of organic poultry according to the Code of Federal Regulations [CFR (Code of Federal Regulations), 2002; Sebranek et al., 2012]. However, in 2016 the National Organic Standards Board approved lactate as an approved substance for use as an antimicrobial agent and pH regulator in organic processing (Favre, 2016). Sodium diacetate was also discussed in this recommendation and determined not to be ancillary and therefore not approved for use.

OPPORTUNITIES FOR *LISTERIA* spp. CONTAMINATION IN POULTRY PROCESSING ENVIRONMENTS

Environments such as those associated with food processing appear to be a natural niche for *Listeria* spp. and this certainly would appear to hold true for conventional poultry processing plants as well (Cox et al., 1997; Lunden et al., 2003; Lianou and Sofos, 2007). In most food sectors, the main contamination sources of *L. monocytogenes* were determined to be equipment, conveyor belts, trays, floors, drains, and workers (Verghese et al., 2011; Barbut, 2016). Surfaces such as slicing machines, packers, and conveyors that are in contact with the food product can become contaminated with persistent *L. monocytogenes* that can be brought into the plant from water sources, workers, or the raw product (Ferreira et al., 2014; Park and Kang, 2014). Once within the processing plant, persistent strains of *L. monocytogenes* can form biofilms, especially in crevasses on the processing machinery of drainage systems (Rørvik et al., 1995; Ferreira et al., 2014). The persistence of *L. monocytogenes* on food contact surfaces has been reviewed by Carpentier and Cerf (2011). Repeated isolation during routine inspection has indicated that in some facilities the pathogen can persist up to a year and potentially longer (Lawrence and Gilmour, 1995; Carpentier

and Cerf, 2011). Rørvik et al. (1995) used multilocus enzyme electrophoresis on isolates of *Listeria* spp. from a smoked salmon slaughterhouse and processing plant. They determined that the same serotype persisted for up to 8 months on surfaces such as the fileting, slicing, and vacuum packaging equipment. These samples were acquired by using cotton swabs on surfaces that are in contact with the product and indicate the need for proper sanitation (Rørvik et al., 1995).

The Food Safety Inspection Service (FSIS) indicate that processing surfaces that have direct contact with the product as well as drains and non-contact surfaces should be swabbed, and *Listeria* presence detected through traditional microbiological techniques or conventional molecular techniques such as polymerase chain reaction (PCR) (Food Safety Inspection Service., 2014). However, Rothrock et al. (2013) found that droplet digital PCR (ddPCR) was able to detect the gene patterns indicative of *L. monocytogenes* in the scalder and chiller water of a poultry processing facility, when conventional plating methods did not detect the pathogen. This may indicate that typically utilized microbiological techniques are insufficient in some processing plants for ensuring product safety and thus more sensitive techniques may be required.

Conventional poultry processing plants have not received as much attention as non-poultry food processing plants for *Listeria* spp. contamination, but the similar environmental conditions would appear to be conducive for the presence of detectable *Listeria* spp. There have been reports that support this. Previous studies have detected the growth of *Listeria* spp. on floors, walls, and drainage of poultry processing facilities (Blackman and Frank, 1996; Verghese et al., 2011). Floor drains in food processing facilities are a particularly important niche for the persistence of *Listeria* spp. and can be a point of contamination in conventional processing plants and possibly in food products (Blackman and Frank, 1996; Berrang et al., 2013). Berrang et al. (2013) constructed model floor drains out of polyvinyl chloride (PVC,) added 650 mL of phosphate buffered saline (PBS) containing 8 log cells/mL of *Listeria innocua* to each model drain and the model floor drains were subsequently allowed to incubate for 2 h at 25°C. *Listeria innocua* has been previously used as a substitute for *L. monocytogenes* in modeling studies as it is largely non-pathogenic, although there have been rare cases in which bacteremia can occur (Perrin et al., 2003; Berrang and Frank, 2012; Milillo et al., 2012a). These inoculated drains were subsequently emptied, rinsed, and placed in an animal house facility. A stainless steel table with 10 chicken breasts was 2.4 m away from the drain and 84 cm from the floor and underneath an inflow air duct (flow rate 5.6 m³/min). Floor drains were then sprayed for 2 s with tap water at 69 kpa. After 10 min, five of the breast meat samples were evaluated for contamination of *L. innocua* caused by airborne contamination in accordance with standard FSIS methods [United States Department of Agriculture (USDA), 2008]. The remaining five breast meat samples were stored for 4 days (5°C) before sampling. A control group was also performed where chicken breast meat samples were left in the room with the contaminated floor drain and no spray was performed. Across five replications ($n = 25$), the mean most probable number per gram of *L. inoccua* on the chicken breast

meat samples was 18.32, which was significantly higher than the control group (0.72). Interestingly the air exposed group had lower *L. inoccua* populations when left in cold storage. This study indicates that drainage pipes and other indirect contact surfaces still have the potential to be reservoirs of pathogens including *Listeria* spp. that can adulterate the finished product.

Less is known about non-conventional poultry processing environment and the occurrence of *Listeria* spp. Although few live bird studies in non-conventional production systems have been examined, there are indications that *Listeria* spp. can occur in birds raised in these environments. *Listeria* spp. were isolated on all 10 pastured poultry farms sampled by Locatelli et al. (2017) and 15% of the fecal samples tested positive for the pathogen. Esteban et al. (2008) found 26.5% of fecal samples from free-range flocks tested positive for *Listeria* spp. In France, 37% of samples from free-range layer hens tested positive for *Listeria* spp. compared to 28% in conventionally-raised caged hens and similar results were found in broilers (Chemaly et al., 2008). The occurrence of *Listeria* spp. in live poultry production has been extensively reviewed by Rothrock et al. (2017).

These higher rates of *Listeria* spp. can potentially be transferred to the processing facility. As such, it is imperative to highlight possible issues and places of potential *Listeria* spp. contact during both conventional and alternative processing of poultry and as well as the survival in/on poultry products that reach the retail environment, and ultimately the consumer. In this review, the similarities and contrasts will be examined between conventional and non-conventional poultry processing and the opportunities for *Listeria* spp. contamination in these respective systems.

One area with a high potential to cause microbial contamination in both conventional and non-conventional poultry processing systems is evisceration due to the removal of the GIT (Lillard, 1989). Mobile poultry processing units typically manually eviscerate the carcass, vs. conventional plants which utilize an automatic system (O'Bryan et al., 2014). Two poultry slaughterhouses in Brazil were sampled for the presence of *Listeria* spp. by Chiarini et al. (2009). While the plants were similar in structure, one processed 130,000 birds a day through automated equipment compared to 70,000 manually processed by the other plant. Over the course of the year, 432 samples per plant were collected from food-contact and non-contact surfaces. Only 1 sample in the automated plant tested positive for *Listeria* spp. prior to evisceration. For post-evisceration, 20.1% of the samples from the automatic evisceration plant tested positive for *L. monocytogenes* while only 16.4% tested positive in the manual evisceration plant. This study did not control for the near doubling of the carcasses processed within the automated plant, but it suggests that manual evisceration may result in lower *Listeria* spp. contamination.

Stainless steel is an ideal surface material used in processing plants because it is easy to clean yet durable, and resistant to chemical deterioration (Ryser and Marth, 2007). It is often the material of choice for both evisceration tables and/or automated machines (New Entry Sustainable Farming Project, 2012). Stainless steel is widely used as a surface material in many environments where there is a risk of bacterial

cross-contamination (Berrang et al., 2000). Most issues in plants arise with improper handling and hygiene during the cleaning of the equipment. *Listeria monocytogenes* has been isolated from the surface of cutting knives and tables in processing plants 24 h after cleaning (Ryser and Marth, 2007). *Listeria monocytogenes* can survive for different periods of time on different metal alloys (Wilks et al., 2006), including alloys often associated with tables, knives, saws, and bird cages. Wilks et al. (2006) conducted research on implications for cross-contamination on metal surfaces which, after achieving a better understanding of these characteristics, could lead to important improvements in public health in food processing, domestic, and healthcare environments. They concluded that the use of copper-based alloys reduced *Listeria* spp. prevalence.

Other environmental factors in the processing plant such as the presence of biofilms may play a role in *L. monocytogenes* persistence. The capability of *L. monocytogenes* to attach to common food contact surfaces such as plastic, rubber, stainless steel, and glass is due to their ability to produce and form biofilms (Wilks et al., 2006; Giaouris et al., 2015). Biofilms, an assemblage of microbial cells that are irreversibly associated with a surface and enclosed in a matrix of primarily polysaccharide material (Manios and Skandamis, 2014), are considered a major problem in the food processing industry. *Listeria monocytogenes* can attach to and survive on various working contact surfaces within these biofilms, and in this situation, they have been shown to be more resistant to biocides which increases the risk of food contamination (Manios and Skandamis, 2014). Out of 40 tested strains of *L. monocytogenes,* all produced biofilms on plastic surfaces immersed in brain heart infusion agar (Stepanović et al., 2004). Of these strains, only 2 produced weak biofilms, as determined by optical density (Stepanović et al., 2004). This may be a concern for MPPUs as *L. monocytogenes* have been demonstrated to adhere to hydrophobic materials, such as plastic, in higher numbers than hydrophilic materials, such as stainless-steel (Cunliffe et al., 1999; Sinde and Carballo, 2000; Donlan, 2002; Stepanović et al., 2004).

The presence of non-*Listeria* microorganisms may also be a factor. For example, *L. monocytogenes* readily colonized *Pseudomonas fluorescens* biofilms (Puga et al., 2018). Biofilms of both species were grown on hydrophilic glass surfaces separately and were also inoculated simultaneously at a concentration of 4 log CFU/mL. After 96-h incubation at 20°C, *L. monocytogenes* concentrations were 1–2 log units higher when grown with *P. fluorescens* and the biofilm density (OD 595 nm) was 1.40 compared to 0.6 for biofilms formed by *L. monocytogenes* alone. This suggests that due to the complex diversity of microbiota in processing facilities, studies should be conducted to investigate and compare the ability to remove mixed biofilms that contain *L. monocytogenes* instead of single species biofilms.

As previously stated, *L. monocytogenes* can replicate on abiotic surfaces present in the food processing environment, but other explanations for *Listeria*'s persistence have also been suggested. Some characteristics of *Listeria* spp. include ability to grow and survive at low temperatures, ability to elicit a survival response and undergo physiological adaptation to nutrient deprivation, competitiveness for nutrients (largely due

to biofilm formation), resistance to cleaning products, random primary colonization, and over time a reintroduction of the same strain (Blackman and Frank, 1996). Biofilms can be difficult to control since they typically form where water is plentiful and cleaning is not performed properly, which can reduce the antimicrobial effects of some products (Verghese et al., 2011); (Manios and Skandamis, 2014).

The ability of biofilms to undergo sanitation efforts and withstand stressful conditions of cleaning is a great concern. Biofilms formed by *Listeria* spp. appear to be more resistant to stress and sanitizing agents than planktonic cells (Blackman and Frank, 1996). Previous studies have shown that biofilm resistance is more prevalent on polyester surfaces, such as conveyor belts, than it is on stainless steel equipment (D'costa et al., 2006; Pan et al., 2006). This suggests that by producing biofilms, *L. monocytogenes* can overcome stresses and antimicrobial interventions resulting in product contamination.

According to Lawrence and Gilmour (1995), lineage I - 4b is the most prominent *L. monocytogenes* serotype in the processing environment (Lee et al., 2018). This serotype is persistent on equipment surfaces, which can result in a possible source of cross-contamination. Pathogens occupying niches in areas such as walls, drains, and floors, which do not have contact with the food product, tend to affect the products produced toward the end of the processing (Lawrence and Gilmour, 1995; Berzins et al., 2009). This may be due to the transfer of the pathogen to workers and their subsequent transferring of the bacterium to the product. Handling of poultry products can play a role in the prevalence of *L. monocytogenes* (Lawrence and Gilmour, 1995). Workers that are constantly handling the carcasses during cutting and evisceration can transfer the bacteria through touching other places in addition to the birds (Brant et al., 1982). *Listeria* spp. can also possibly be transferred through poor hygiene of carcass handlers. In an investigation by Kerr et al. (1993), 99 workers engaged in food production and retailing were sampled, and analysis revealed 12% of food workers harbored *Listeria* spp. Of the 12% that were contaminated, 7% harbored *L. monocytogenes* (Kerr et al., 1993). In MPPUs, fewer workers are present with some only requiring one operator (O'Bryan et al., 2014; Mancinelli et al., 2018). As such, hygiene is essential, lest the entire processing run can be potentially contaminated. Previous research has shown that hand washing is not an efficient method of bacterial removal when the density of bacteria is extensive, and those that work in the processing area with direct contact with poultry products are significantly more likely to carry *Listeria* spp. than their non-processing plant counterparts (Snyder, 2001).

During poultry processing, *Listeria* spp. can contaminate different areas of the meat based on preference and processing condition. Typically, contamination occurs through transmission from the processing facilities and equipment (Blackman and Frank, 1996). Due to the ubiquitous nature of *L. monocytogenes*, they can be isolated from both fresh and frozen carcasses (Van Nierop et al., 2005). Previous studies conducted by Barbalho et al. (2005) isolated *L. innocua* from carcass samples at all steps of the processing line, including bleeding, scalding, de-feathering, evisceration, and packaging. *Listeria monocytogenes* were present only in RTE packaged meats for retail. In another

study, Berrang et al. (2000) found that *L. monocytogenes* was 6% less prevalent on eviscerated post-chill chicken carcasses than in the pre-scalding stage and they suggested that it may be possible to prevent cross-contamination of poultry products in the plant by removal or replacement of processing equipment. Processing lines, environment, personnel, raw materials, and RTE products were studied for the presence of *L. monocytogenes* after sanitization and after the process had been running for 2 h. *L. monocytogenes* were detected in samples taken from drains and floors in 11 of the 13 conventional processing plants (Gudbjörnsdóttir et al., 2004). In the raw poultry products, 22% were contaminated with *Listeria* spp. and the incidence rates within the final RTE poultry meats were 2.2%.

In an effort to improve surveillance for *Listeria* spp. in processing plants, pulse-field gel electrophoresis (PFGE) and other molecular techniques have been applied (Lopez-Valladares et al., 2017). Fox et al. (2015) evaluated 6,785 samples in a poultry processing facility over 23 months and found that 109 of these samples contained *Listeria* spp. as detected by PFGE. The frequency of *L. monocytogenes* was relatively low at 0.94%. Of the 20 PFGE patterns identified, only two (LmT03 and LmT22) were found to persist from ingredients to the food preparation areas. Because few PFGE sub-typing profiles came from the ingredients, it must be assumed that the *Listeria* was previously introduced or brought in through other avenues such as workers, water, or unsampled sites. By including this data when surveying *Listeria* spp., contamination sources can be identified and appropriate control measures applied.

Listeria spp. Prevalence in Poultry Retail Products

Listeria contamination occurs in raw retail poultry meat. Previous studies show *L. monocytogenes* to be present in raw poultry in supermarkets (Lawrence and Gilmour, 1994; Goh et al., 2012). Elmali et al. (2015) sampled 120 broiler wings from 5 different stores in Turkey and found 47% of broiler wings contained *Listeria* spp., with 54 of the 57 isolates being *L. monocytogenes*. These chicken wings were acquired during each month over the course of the 1 year study to account for potential batch contamination. This study tested for prevalence by adding 25 g of wing to 225 mL of ONE-broth *Listeria* (Oxoid, Basingstoke, Hampshire, England) and after 24-h incubation at 30°C was inoculated on Brilliance *Listeria* Agar plates (Oxoid) and incubated at 37°C for 24 h. Green-blue colonies were considered indicative of *Listeria* spp. and *L. monocytogenes* was determined through PCR amplification of the *hlyA* gene. This study suggests that poultry may be contaminated due to poor processing techniques, along with other factors such as temperature and water activity (Elmali et al., 2015). Raw poultry is a favorable environment for *L. monocytogenes*, and once it contaminates the meat in the processing environment, there is a high chance it will survive through to the retail environment and ultimately to the consumer (Ceylan et al., 2008). This is why it is critical that the proper processing techniques as stated earlier are followed explicitly.

The prevalence of *L. monocytogenes* in RTE poultry products is also of great public health concern. The extended distribution throughout the environment and the psychrotrophic nature of *L. monocytogenes* appear to be the main cause of the high prevalence in different types of refrigerated RTE products (Ryser and Marth, 2007). The increase of RTE consumption, due to difficulties to controlling temperature in the global trade distribution of these products as well as changes in consumer lifestyles could be some of the reasons for the increase of listeriosis outbreaks (Garrido et al., 2010). Premade foods, such as poultry delicatessen meats, are common in average households because they are an efficient time saver, which means packaged meats are at the forefront for consumers. The safety of packaged poultry is a threat because *L. monocytogenes* can grow even at refrigeration temperatures (Ryser and Marth, 2007). This can be a potential problem since the shelf life of meat and poultry packaged in the CO_2 mixture is 45–70 days longer than that of meat packaged in the commonly used atmospheres with high oxygen (O_2) (Sorheim et al., 1997). In the literature there is controversy over the effectiveness of controlling *Listeria* through modified atmospheric packaging as it is a facultative anaerobic organism and can survive in a wide range of CO_2, O_2, and N_2 concentrations (Jacxsens et al., 1999; Heinrich et al., 2016).

Previous reports have shown an increase in the prevalence of *L. monocytogenes* in RTE, vacuum packaged, sliced meat and turkey products where 95% of all *L. monocytogenes* belonged to Lineage II, serotype 1/2a, with the remaining 5% varying between serotypes 1/2b, 3b, and 4b (Berzins et al., 2009). Kramarenko et al. (2013) reported that 93% of all *L. monocytogenes* isolates obtained from meat and chicken products belonged to serotype 1/2a and 1/2c. Therefore, this suggests that variations between stress and exposure influence which lineage, serotype, and strain is ultimately responsible for the contamination. Novel identification tools such as whole genome sequencing based on single-nucleotide polymorphism phylogenetics may be used to further identify and track the specific serotype and strain of *Listeria* within food systems (Stasiewicz et al., 2015).

In November 2016, National Steak and Poultry of Oklahoma recalled ~17,439 pounds (7,910.33 kg) of RTE chicken products because of possible undercooking. Treatment with high temperatures during cooking is a means to reduce and possibly eliminate the number of potential pathogens. Without proper temperature treatment and handling, illness can occur after ingesting pathogen contaminated food [United States Department of Agriculture (USDA), 2016]. Previous studies have demonstrated that packaging and its effect on CO_2 levels contribute to the prevalence of *Listeria* spp. (Zhu et al., 2005). Simmons et al. (2014) found of 4,503 samples collected from 28 different food contact sites in 30 delis that 9.5% of samples were positive for *Listeria* spp. Approximately 450 *L. monocytogenes* isolates were identified using PFGE and 12 of the 30 delis contained PFGE types that were persistent across at least three of the six sampling points during the 6 month study. Irradiation is an effective post-packaging intervention technique used to eliminate *L. monocytogenes* from contaminated RTE meat products; however, it can influence product quality by altering the color, flavor and can induce lipid oxidation (Zhu et al., 2005).

Low dosage irradiation is recommended as a processing aid for RTE products. Ultraviolet (UV)-C radiation, which emits wavelengths of 220–300 nm, has been approved by the FDA on food products to control surface microorganisms [United States Food Drug Administration (FDA), 2007]. Chicken breast meat samples inoculated with 6 log CFU/g *L. monocytogenes* were reduced by 1.29 log CFU/g after receiving a dose of 5 kJ/m^2 (Chun et al., 2010). After 6 days of cold storage, populations of *L. monocytogenes* in the 5 kJ/m^2 treatment group were 4.6 log CFU/g compared to 5.7 log CFU/g in the control group. In the 0.5 kJ/m^2 treatment group a 0.5 log CFU/g difference was observed by the end of the 6-day storage compared to the control. This indicates that UV-C radiation may be a suitable hurdle to reducing *L. monocytogenes*. However, it is important to note that high levels of inoculum may not be an accurate representation of real world contamination, and may overstate the effectiveness of treatments (Rodriguez et al., 2007).

Miranda et al. (2008) collected fifty-five 25 g samples of organic drumsticks from 5 different supermarkets in northwest Spain. Samples were stomached, homogenized in peptone and investigated for *L. monocytogenes*. Of the 55 organic drumstick samples, 27 tested positive for *L. monocytogenes* with an average log colony forming unit (CFU)/g count of 0.942 (Miranda et al., 2008). Additionally, 61 conventionally processed drumsticks samples were collected, and 36 isolates of *L. monocytogenes* were acquired with an average 0.785 log CFU/g. The isolation rates and CFU counts between conventional and organically processed drumsticks were statistically similar ($P > 0.05$). These isolates were subsequently tested against chloramphenicol, doxycycline, erythromycin, gentamicin, sulfisoxazole, and vancomycin. Doxycycline resistance was significantly higher in isolates of conventionally raised poultry ($P < 0.05$) and all other resistances were not significantly different between the two groups. To the best of our knowledge, no study has currently evaluated *Listeria* spp. contamination of MPPUs or other alternative processing facilities. However, Rothrock et al. (2016b) did investigate multi drug resistance (MDR) isolates in pasture flocks and found comparable rates of MDR *Listeria* spp. in flock processed by a USDA inspected facility and on-farm processing. These isolates were acquired from feces, soil, ceca, whole carcass rinses and on the final product. However, understanding potential control measures is necessary to proposing solutions once persistence and prevalence information becomes readily available.

POTENTIAL CONTROL MEASURES FOR *LISTERIA* IN POULTRY PROCESSING

Extensive work has been done with administration of antimicrobials in conventional food products and processing environments, most of which is applicable to poultry products as well (Xi et al., 2012). *Listeria* spp. contamination may be mitigated through staff training and production procedures (Hicks et al., 2004). In RTE meats and poultry, Hazard Analysis and Critical Control Points (HACCP) plans are applied to reduce chemical and biological adulterants (Barbut, 2015). In poultry slaughtering control points can include the scalder, evisceration, final wash, chilling, and storage for further processing (Tompkin,

1994; Barbut, 2015). The further processing critical control points potentially include receiving, weighing, cooking, chilling, emulsifing, and packaging in RTE poultry production (Barbut, 2015). The HACCP model was extensively detailed in Barbut (2015) and will not be reviewed further.

To reduce bacterial loads chemical sanitizers such as chlorine and PAA can be applied in poultry processing (Micciche et al., 2018b). Chlorine (100 μg/ml) and PAA (80 μg/mL) were added to a 1 L dip and 10 g of lettuce inoculated with 5 log CFU/g *L. monocytogenes* were subjected to the antimicrobial treatment for 15 s (Beuchat et al., 2004). Chlorine and PAA reduced recoverable *L. monocytogenes* by 3 and 4 log CFU/g, respectively, however lettuce and other non-poultry media may not be representative of *Listeria* contamination of poultry products. Despite the effectiveness of sanitation, some concerns do exist (Ibusquiza et al., 2011; Olaimat et al., 2018). For example, due to exposure of various antimicrobials and treatment of antibiotics prior to processing, antibiotic resistance frequency amongst *L. monocytogenes* isolates can increase. Strains that are resistant can proliferate in the processing environment, which increases the risk for consumers (Kerr et al., 1993; Jarvis et al., 2015). The addition of antimicrobials can limit the growth of *L. monocytogenes*; however, antimicrobials can reduce product quality (Ryser and Marth, 2007). *Listeria* spp. possess the ability to resist antimicrobials in RTE poultry since antimicrobials do not fully destroy the pathogen (Zhu et al., 2005). Others have determined that *L. monocytogenes* can be reduced with lactates and diacetates, which are two of the most common and effective of antimicrobials used in RTE poultry meat (Bedie et al., 2001; Barmpalia et al., 2004). Lactates and diacetates, when added in the formulation as growth inhibitors in the immediate packaging material, have been demonstrated to be effective in the control of *L. monocytogenes* in RTE poultry [U.S. Department of Agriculture Food Safety Inspection Service (USDA-FSIS), 2006; Knight et al., 2007]. Antimicrobials can be added to the product during formulation, the finished product or to the packaging material. Processing facilities should use antimicrobial agents that have been approved by the FDA and USDA-FSIS for processed RTE meat and poultry products.

Non-conventional poultry processing represents additional challenges due to restrictions on the types of antimicrobials that can be added. These restrictions can occur due to cost, the use of organic labeling, and transportability in the case of MPPUs (Fanatico, 2003; Micciche et al., 2018a; National Organic Program, 2018). General sanitation options typically include chemical sanitizers such as chlorine, PAA, or ozone, which may be used in organic processing (Northcutt and Jones, 2004; Mahapatra et al., 2005; O'Bryan et al., 2014). However, due to the need for on-site generation, ozone is not applicable for MPPUs (Garud et al., 2019). Chlorine can produce gaseous by-products, and PAA is moderately volatile, which can be hazardous to operators in small closed areas such as closed-air MPPUs [Pedersen et al., 2013; Occupational Safety Health Administration (OSHA), 2017; Micciche et al., 2018b]. As such, alternatives such as essential oils, bacteriocins, and phages should be explored and are discussed in detail for their application against *L. monocytogenes*.

One proposed alternative biological method is the use of phage therapy (Micciche et al., 2018b). Phages are the most abundant organisms on earth and are viruses that infect bacteria for propagation (Pérez Pulido et al., 2016). Phages operate either under a lytic or lysogenic lifecycle, where, in the latter, the prophage may insert itself into the genome of the host and propagate over generations until an environmental stressor triggers unfavorable conditions resulting in a shift to the lytic cycle and lysis of the cell (Gray et al., 2018). In the lytic life cycle, phages, upon attachment and entry to the cell, propagate using host machinery and lyse the cell immediately. They are suitable as a biocontrol agent unlike their lysogenic counterparts due to host cell integration of the prophage (Salmond and Fineran, 2015). The underlying mechanisms of phage therapy are discussed in Gray et al. (2018). Phages can specifically infect species or strains of bacteria depending on their host range, with those selected for phage therapy replicating within their bacterial hosts and lyse the cell afterward (Shors, 2001).

Phage therapy is considered organic, and the FDA has approved phage cocktail products for the reduction of *Listeria* spp. on meat and poultry products during processing or in the final product (Sulakvelidze, 2011; Ricke et al., 2012; OMRI., 2019). In 2013, Chibeu et al. (2013) evaluated LISTEX^{TM} P100, a proprietary phage cocktail, against a four-strain blend of *L. monocytogenes* at a concentration of 3 log CFU/cm^2 on RTE roast beef and turkey. Both the bacteria and phage treatment were added to the product prior to vacuum sealing. After a 28-day storage period at 4°C, *L. monocytogenes* numbers were ~2 log CFU/cm^2 lower relative to the control. Initial reductions of 1.5 log CFU/cm^2 were observed for phage therapy on the cooked turkey. When using LISTEX^{TM} P100 on biofilms formed on stainless steel, 3.5–5.4 log CFU/cm^2 reductions were observed depending on which of the 21 strains were tested (Soni and Nannapaneni, 2010; Gray et al., 2018). Similar reductions were observed by (Monta-ez-Izquierdo et al., 2012), which also tested LISTEX^{TM} P100 biofilms *L. monocytogenes* on stainless steel. LISTEX^{TM} P100 was also applied for 24 h on *L. monocytogenes* biofilms that grew on stainless steel coupons and transferred to Palcam agar plates (Iacumin et al., 2016). No growth of *L. monocytogenes* (limit 1 log CFU/cm^2) was observed when treated with phages. Sadekuzzaman et al. (2017) tested LISTSHIELD^{TM} on *L. monocytogenes* biofilms and found its application for 2 h reduced bacterial concentrations by 2 log CFU/cm^2 on stainless steel biofilms and 1 log CFU/cm^2 on biofilms formed on rubber. These studies did not investigate biofilms formed on plastic. Future studies should investigate the formation of *Listeria*-based biofilms on equipment utilized in conventional and MPPU and evaluate if these alternative antimicrobials can be utilized effectively. Currently, phage therapy may not be applicable to small scale processing facilities due to its high cost (Micciche et al., 2018a). But due to their persistence within processing waters and the potential for new innovations to lower initial capital costs it is possible that small scale poultry processors may find this remediation method viable.

To attract consumers concerned with chemical additives, clean label ingredients (ingredients with common consumer use) have been suggested (Van Loo et al., 2011; Badvela et al., 2016). Dry vinegar with a sodium base was tested on turkey to control *L. monocytogenes* in cold storage (Badvela et al., 2016). Turkey breast meat samples were inoculated with 5 log CFU/100 g of a five-strain *L. monocytogenes* cocktail. Breast meat samples were subjected to 0.4, 0.6, and 0.8%, sodium buffered dry vinegar and placed into cold storage 4°C. *Listeria* concentrations were then measured once a week for 12 weeks. The cocktail consisted of *L. monocytogenes* 101 (serotype 4b), *L. monocytogenes* 108 (serotype 1/2a), *L. monocytogenes* 310 (serotype 4b), FSL-C1-109 (serotype 4b), and V7 (serotype 1/2a). A second cocktail using different strains was also prepared but no significant differences were observed between the inocula in this study. By the end of the study, *L. monocytogenes* counts had risen by 1.89 log CFU/100 g, while there were no significant changes in concentration in the 0.6% treatment group, and a 0.5 log CFU/100 g reduction was observed in the 0.8% treatment group. Similar results were found when using a phosphate base instead of sodium. In the no treatment control, bacterial counts had risen by 6 log CFU/100 g. This suggest s that while dry vinegar may not have significant bactericidal properties against *L. monocytogenes,* it could still be used to prevent *Listeria* spp. growth.

Another product that may control for *Listeria* and has potential applications in organic production and MPPU is the use of lactate and lauricidin (glycerol monolauric acid) (Anang et al., 2007). Lauricidin is found within coconut oil and is considered organic (Hegde, 2006; National Organic Program, 2018). Raw chicken breast meat samples were inoculated with 7–8 log CFU/g of *L. monocytogenes* (L55) and subjected to a 10, 20, or 30 min submersion in a 0.5, 1, 1.5, and 2% solution of either lactic acid or lauricidin (Anang et al., 2007). While reductions ranged over time and concentration, a 2.68–2.90 log CFU/g reduction was observed for the lauricidin treatments, and a 0.95–1.96 log CFU/g reduction was observed for the lactic acid treatments. When stored for 14 days at 4°C, all lauricidin and lactic acid treatments produced significant reductions in *L. monocytogenes* concentrations compared to the control. These treatments were also tested on *Salmonella* Enteritidis and *Escherichia coli* O157:H7 and reduced bacterial concentrations significantly by day 14 at all concentrations. It is important to note the ability of lactic acid to impact other bacteria populations, because while lactic acid has not been tested against *L. monocytogenes* in MPPUs, it has been tested in that environment as an alternative to chlorine (Killinger et al., 2010). After evisceration and a water spray wash, carcasses were immersed in a no-treatment control, 2% lactic acid solution, or 50–100 ppm chlorine solution for 3 min. Twenty carcasses were utilized per treatment, and the experiment was replicated three times. Chlorine significantly reduced aerobic plate counts by 0.4 log CFU/carcass and coliform counts by 0.21 log CFU/carcass. Lactic acid reduced aerobic plate counts by 2 log CFU/carcass and coliform counts to below 0.5 log CFU/carcass. A future study utilizing lactic acid as a sanitizer against *L. monocytogenes* in an MPPU and conventional processing plant should be performed to determine its effectiveness against the pathogen in a processing environment.

Another suggested group of sanitizers for alternative poultry processing are essential oils (Micciche et al., 2018a). Essential oils (EOs) are derived from botanicals or phytobiotics and there are over 3,000 known EOs with ∼300 being commercially relevant (Bakkali et al., 2008; Diaz-Sanchez et al., 2015; Liu et al., 2017). Essential oils, such as thyme, eugenol, and oregano, have the potential of producing off flavors when utilized in sufficient quantities (Calo et al., 2015). This is a detriment to large scale poultry processing where meat product sensory consistency is critical (Micciche et al., 2018a). However, provided the flavor is palatable, small-scale facilities such as MPPUs have a unique opportunity to market their product by promoting the EOs flavor. For instance, the application of 0.1% oregano oil improved the flavor and taste of chicken breast when utilized in the packaging (Chouliara et al., 2007). While no study has evaluated the impact of sensory characteristics of EOs when applied in the chiller tank or wash water, these products may confer advantages to taste and odor if used in appropriate concentrations.

The use of EOs against foodborne pathogens has been reviewed by Diaz-Sanchez et al. (2015), Micciche et al. (2018a), Calo et al. (2015), and O'Bryan et al. (2015). Friedman et al. (2002) evaluated 96 EOs and 23 oil compounds against *Campylobacter jejuni, Escherichia coli* O157:H7, *L. monocytogenes* RM2199 and RM2388, and *Salmonella enterica* at a concentration of 4 log CFU/mL. Essential oils were applied in PBS for 60 min at 37°C. Bactericidal activity that resulted in 50% CFU decrease relative to the control was determined for each concentration of EO. Cinnamaldehyde (0.019%), gardenia (0.057%), cedarwood (0.067%), bay leaf (0.070%), and clove bud (0.074%) were the most effective oils against *L. monocytogenes* RM2199. Cinnamaldehyde (0.019%), cedarwood (0.028%), patchouli (0.029%), gardenia (0.038%), and orange sweet (0.040%) were the most effective oils against *L. monocytogenes* RM2388. Broad spectrum screening for antimicrobial activity of EOs against *L. monocytogenes* was also performed by Oussalah et al. (2007). The minimum inhibitory concentration (MIC) for 28 EOs was calculated against *L. monocytogenes* 2812 (1/2a) at an initial concentration of 7 log CFU/mL. This study indicated that cinnamaldehyde (0.05%), along with savory (0.1%) and EOs derived from several strains of oregano (0.025, 0.05, 0.1%), were among the most effective in reducing *L. monocytogenes*.

Essential oils have also been found to reduce *L. monocytogenes* in the finished poultry product (Mytle et al., 2006). Chicken frankfurters were inoculated 5 log CFU/g of *L. monocytogenes* and subjected to 0, 1, or 2% of clove oil for 30 min and stored at 4°C for 14 days. Seven *L. monocytogenes* strains were evaluated in this study all from serotype 4b or 1/2a. While impact varied from strain to strain all concentrations of pathogen populations were reduced by the application of 1–2% clove oil compared to the controls. For example, *L. monocytogenes* G3982 (4b) concentrations were 3.7 log, 1.1 log, and 0.2 log CFU/g for the 0, 1, and 2% clove oil treatments, respectively. Future studies will need to evaluate the impact that other EOs have on *L. monocytogenes* on chicken products and within the processing plant.

Other natural antimicrobials have been investigated for their use against *Listeria* spp. such as liquid smoke (Babu et al., 2013; Lingbeck et al., 2014). Liquid smoke is produced by condensing smoke created by burning flammable materials and subsequently removing carcinogenic polycyclic aromatic hydrocarbons (Lingbeck et al., 2014). Extract of organic pecan shells was tested on seven *Listeria* strains by Babu et al. (2013). A proprietary solvent-free extract system was applied and volatile gaseous were condensed into liquid smoke. They found that pecan shell extract exhibited a minimum inhibitory concentration (MIC) of 0.375% on the cocktail of all seven strains and the highest MIC was 6% (Lm 190, 1/2a). When inoculated chicken skin was incubated for 30 min with 0.75% pecan extract, a significant 1 log CFU/cm^2 reduction was observed compared to the control which had a concentration of 6.5 log CFU/cm^2. Sensory analysis was not reported in this study. This reduction is observed in other meat commodities as well. For example, two liquid wood smoke preparations were found to reduce *L. monocytogenes* in rainbow trout below the limit of detection from an initial concentration of 2.5 log CFU/g (Lingbeck et al., 2014). This was accomplished by dipping the inoculated trout in liquid smoke concentrate for 1 min. However, a third liquid wood smoke preparation did not reduce *Listeria* spp. concentrations. Differences between the smoke preparations and the concentration used were not stated. Future studies investigating liquid smoke would be valuable as understanding how liquid smoke reduced microbial concentrations is not well established.

Bacteriocins, which are also natural alternative antimicrobials have been found to be effective against *Listeria* spp. (Bruno and Montville, 1993). Bacteriocins are produced by microorganisms to inhibit similar bacteria and operate by disrupting the cytoplasmic membrane and destroying the electrostatic potential of bacteria, notably Gram-positive species (Ruhr and Sahl, 1985; Bruno and Montville, 1993; Sirsat et al., 2009). Nisin, produced by *Lactococcus lactis*, is the only generally recognized as safe (GRAS) bacteriocin [Shin et al., 2016; United States Food Drug Administration (FDA), 2018]. The addition of 2.5 mg/L of nisin to scalder water in turkey processing reduce *L. monocytogenes* by 1 log CFU/g compared to the 5.5 Log CFU/mL control (Mahadeo and Tatini, 1994). When used in conjunction with heat (52°C for 3 min) a 2 log CFU/mL reduction was observed and after 48 h refrigeration no *Listeria* was detected. In smoked pork sausage inoculated with 3.5 log CFU/cm^2 *L. monocytogenes* were stored at 10°C for 48 days and 1 log CFU/cm^2 reductions were observed on products first immersed in 5,000 international units of nisin/mL for 2 min (Geornaras et al., 2006). On chicken sausages inoculated with 2 log CFU/g *L. monocytogenes*, the application of 12.5 μg/g of nisin for 15 min reduced concentrations below the limit of detection (Sant'Anna et al., 2013). A *Bacillus* sp. bacteriocin (P34) at a concentration of 128 μg/g reduced *L. monocytogenes* in chicken sausages by 1 log CFU/g. The antimicrobial effect of bacteriocins may be valuable in a multiple hurdles approach but because they are relatively costly to purify, they may not be viable for small scale poultry processing (Davidson et al., 2005; Fahim et al., 2016). Furthermore, due to the presence of lipids, proteins, and proteolytic

enzymes, constant application of bacteriocins may be necessary as these can interact with the molecule (Bradshaw, 2003; Mahapatra et al., 2005).

CONCLUSIONS

Listeria monocytogenes is a foodborne pathogen of significance that has been isolated from conventional poultry as well as pasture and free-range poultry flocks, and on organic poultry products. The pathogen may persist in conventional processing plants due to biofilm formation. Because of the variation of materials, such as plastic chiller containers, biofilms may form differently in MPPU's and will need to be evaluated independently. The persistence of *L. monocytogenes* is a concern not only because of cross-contamination events, but because it can result in an increase in antimicrobial resistance. To remove these biofilms, however, appropriate sanitizers and antimicrobials are needed that may not be approved for organic processing (McDonnell et al., 2013). As a consequence, effective sanitizers approved for organic processing must be evaluated.

The presence of *L. monocytogenes* has not been reported in MPPU's, due in part to inspection exemptions. While this is due to the low number of birds processed, human illness can be spread particularly to those that are immunocompromised.

Furthermore, because of the potential biofilms, antimicrobial resistance of approved organic sanitizers may be acquired. This can be of particular concern as waste from MPPU's may be discarded on gardens or near rivers and streams. Future studies will need to evaluate if *Listeria* spp. contamination is a concern for these alternative processing units in order to make an appropriate suggestion of which sanitizers may be needed to ensure public health.

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