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Prevalence of selected foodborne pathogens in the processed meat products from Durban and their growth after treatment with vinegar and lemon juice

*Ijabdeniyi, O. A, Naraindath, K. and Ajayeoba, T. A.

Department of Food Science and Biotechnology, Durban University of Technology, Durban, South Africa

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Abstract

The present work determines the prevalence of selected foodborne pathogens in ready-to-eat meat (RTEM) obtained from household and retail locations, and evaluates the effect of vinegar, lemon juice, and combined treatments on the survival of *Listeria monocytogenes* and *Escherichia coli* O157:H7 at different storage temperatures. The pathogens were identified using standard methods and the effects of treatments were evaluated on salami after 144 h. The results obtained from 60 RTEM samples showed a higher prevalence in the household samples than the retail samples, although it was not significantly different ($p > 0.05$). *Staphylococcus aureus* recorded the highest prevalence. Furthermore, 60% of beef sausage samples were positive for *E. coli*, 50% of salami samples were positive for *L. monocytogenes*, while *Salmonella* was lower in all the RTEM categories. *16s rRNA* identified some of the microorganisms such as *Enterobacter cloacae*, *Citrobacter freundii*, *Lelliottia amnigena*, and *L. monocytogenes*. The combined treatment had a higher effect than the single treatment with a 5-log reduction of pathogens; however, the protective effect diminished at storage temperature of more than 4°C. The findings show a higher efficacy of treating foodborne pathogens with combined treatment.

Introduction

Foodborne pathogens are an emerging problem involving a wide spectrum of illnesses caused by bacteria, viruses, parasites, and fungi (Lee *et al.*, 2014). The incidence of foodborne illness may result from the consumption of ready-to-eat (RTE) meals (Christison *et al.*, 2008). Bacterial pathogens associated with RTE food products are *Escherichia coli* O157:H7, *Salmonella enterica* serovar *typhimurium*, and *Listeria monocytogenes*, which result in major disease outbreaks and product recall (CDC, 2014). Processed meat products, especially ham, salami, and bacon are widely sold in delicatessens and consumed in different homes. The handling procedures such as cutting, slicing, and repackaging in small portions, pieces, or slices are considered critical in providing excellent conditions for further contamination, growth, and survival of these pathogens (Ferrentino *et al.*, 2015). Moreover, RTE meat products have been identified as high-risk products, as they are highly perishable and easily contaminated (FSIS, 2010).

One in ten people would fall ill after eating contaminated food, and about 420,000 people die worldwide, as a result, every year (WHO, 2015). In addition, African region along with South-East Asian region have the highest burden of foodborne illnesses. The foodborne illnesses in Africa, including South Africa, have increased because there is no adequate surveillance system or coordinated structure to investigate foodborne outbreaks in RTM food products. Furthermore, there is a high risk of occurrence of foodborne pathogen in South African food processing facilities due to the lack of multiple preservation barriers, storage facilities, rapid detection assays, and poor understanding of the growth mechanism of microorganisms, especially the lack of knowledge about intricate bacterial foodborne pathogens (Gedela, 2007). As of April 2018, a total of

*Corresponding author.
Email: oluwatosini@dut.ac.za

1019 laboratory-confirmed listeriosis cases have been reported to the National Institute for Communicable Diseases (NICD) since January 01, 2017; and 200 deaths have been recorded (NICD, 2018). Polony (a type of processed meat) produced by Enterprise Foods, a division of Tiger Brands, was implicated for the disease.

Household food sanitisers such as vinegar and lemon have been used to reduce microbial load and to inactivate the pathogens contaminating food products (Jans *et al.*, 2017). Various researchers have reported that the use of lemon juice and vinegar could reduce and inactivate *E. coli* O157:H7 and *Salmonella* in a variety of meat products (Li *et al.*, 2012; Chen *et al.*, 2017). However, there is limited information on the effect of combined treatment of lemon and vinegar on the survival of these pathogens. Furthermore, limited work has been done in Africa regarding the safety of processed meat products. The present work thus aimed to determine the prevalence of these foodborne pathogens in processed meat products sold at selected locations and household meat products in Durban metropolis and evaluate the effect of vinegar and lemon juice on the survival of *L. monocytogenes* and *E. coli* O157:H7 at different storage temperatures.

Materials and methods

Sample collection

A total of 60 commercial samples of three different categories (20 ham, 20 salami, 20 bacon) were collected randomly from twenty Durban households (30 samples) with prior consent and retail outlets (30 samples) using sterile LDPE Ziploc bags. The intact samples were stored at -18°C to prevent the growth of the bacteria, and analysed within 24 h. These samples were examined for the presence of aerobic and anaerobic spore-forming bacteria; *Staphylococcus aureus*, *L. monocytogenes*, *Salmonella* spp., and *E. coli* using the conventional method previously described by Ijabadeniyi and Pillay (2017). Fresh lemons and vinegar were purchased from Shoprite outlet, Durban, South Africa.

Media, reagents, and test pathogens

All media and reagents used were purchased from Oxoid (UK) and Sigma Aldrich (US). The test microorganisms (*E. coli* O157:H7 ATCC 43888 and *L. monocytogenes* ATCC 7644) were obtained from the Food Microbiology Laboratory, Durban University of Technology, Steve Biko Campus, South Africa.

DNA sequencing of isolates

Six isolates were randomly chosen from both retail and household isolates and the *16s* target regions of DNA of presumptive positive isolates were identified and extracted using the Quick-DNA™ Fungal/Bacterial Miniprep Kit (Zymo Research, Catalogue No. D6005). The extracted DNA was amplified using the primer 16S–27F 5'AGAGTTTGATCMTGGCTCAG 3' and 16S–1492R 5' CGGTTACCTTGTTACGACTT 3' according to Lane *et al.* (1991). The extracted fragments were sequenced (Nimagen, Brilliant Dye™ Terminator Cycle Sequencing Kit V3.1, BRD3–100/1000), purified (Zymo Research, ZR–96 DNA Sequencing Clean-up Kit™, Catalogue No. D4050), and analysed on an ABI 3500xl Genetic Analyser (Applied Biosystems, ThermoFisher Scientific). Purified sequences were analysed using a CLC Main Workbench 7 followed by a BLAST search as described by Altschul *et al.* (1997).

Preparation of antimicrobial treatment and bacterial analysis of the samples

Lemon juice was extracted from fresh lemon with a juice extractor (JE2400BD, Canada) while pasteurised grape vinegar was used directly. A 2% treatment solution of each of fresh lemon juice, vinegar, and a mixture of vinegar and lemon juice (1:1) were prepared aseptically before use. Moreover, pathogen-free salami samples were purchased from a retail food store to conduct the simulative study. This sample was stored at -18°C after confirming the absence of the test microorganisms (*L. monocytogenes* and *E. coli* O157:H7).

Experimental design

The experimental design followed 2 × 3 × 2 factorial combinations. The effect of three factors: the type of microorganism (*L. monocytogenes* and *E. coli* O157:H7), form of treatments [fresh lemon juice, vinegar, fresh lemon juice + vinegar (1:1 v/v)], and storage temperatures (4°C and 7°C) were evaluated by bioassay of salami cold cuts on daily basis for a period of 6 days. All experiments were carried out in triplicate.

Bioassay of antimicrobial treatments in broth solutions and salami

The inoculum preparation and artificial contamination of test pathogens (*L. monocytogenes* ATCC 7644 and *E. coli* O157:H7 ATCC 43888) was carried out to evaluate their survival on salami according to the method described by Ijabadeniyi and Pillay (2017). A pathogen-free salami sample was used as a control. The effect of different antimicrobial treatments on the pathogens was assessed using

a broth system following the modified method of Leuschner and Zamparini (2002). The salami cold cut samples were prepared following the modified method of Syne *et al.* (2013). Briefly, salami cold cuts were ground in a benchtop blender for approximately 2 min, divided into 25 g portions and mixed with an appropriate quantity of antimicrobial solution (lemon juice, vinegar, and a mixture of lemon juice and vinegar). Each portion was placed into 14 × 20 cm plastic bags and flattened to 5 mm thickness. The plastic bags were heat-sealed and placed into a water bath at 50°C for 30 min before overnight refrigeration (4°C). The treated samples were stored at 4°C and 7°C in the respective refrigerators and taken out after every 24 h to determine the growth of the pathogens over a period of 144 h.

Statistical analysis

The data obtained on the prevalence of pathogens from different locations, RTE meat categories, and treatments of the microorganisms with antimicrobials were subjected to the analysis of variance with SPSS 21.0 version. Prevalence was considered as statistically significant at $p < 0.05$. Duncan's multiple range tests was used to determine the differences between the means.

Results and discussion

Isolation and identification of the microorganisms

Foodborne disease outbreaks are found to be associated with various foodborne pathogens, which are linked with ready-to-eat (RTE) food products (Christison *et al.*, 2008). All the 60 processed RTE meat samples purchased from different retail stores and obtained from different households within Durban metropolis showed contamination of *S. aureus*, aerobic spore-formers, and anaerobic spore-formers. Although the log CFU/g counts observed in the household samples were generally higher, they were not, however, significantly different ($p > 0.05$) from the retail samples (Table 1). Even though the results fall within the acceptable limits according to the South African commission regulation on microbiological criteria for foodstuffs (Turner *et al.*, 2000), the presence of aerobic and anaerobic microorganisms indicates poor quality of these RTE meat products and also cross-contamination due to inadequate handling and/or storage after processing (Syne *et al.*, 2013). Aerobic and anaerobic spore formers, such as *Bacillus* spp. and *Clostridium botulinum*, are the sources of concern because the spores enable the microorganisms to be resistant to heating, freezing, chemicals, and other treatments given to RTE meat products during processing and preparation. This, in turn, decreases the safety and shelf life of RTE meat products (Norashikin *et al.*, 2018). Almanza *et al.* (2007) also reported a lower log CFU/g counts in retail processed meat products than household samples. The higher log CFU/g counts in household samples may be attributed to improper storage conditions at household levels, while the lower counts in retail samples could be due to the use of better refrigeration temperature as well as the use of improved sanitary practices among the retailers (Mani-López *et al.*, 2012).

Figure 1 shows the distribution of food-borne pathogens in the 60 RTE meat samples collected from Durban metropolis. Generally, a higher prevalence of foodborne pathogens was observed in the household RTE meat samples than the retail samples. This higher occurrence of pathogens in the household meat products may be due to the increased survival of enteric pathogens in the RTE meat product, discharge of aerosols, or unhygienic condition of handling and/or abuse of storage temperature in households (Magwedere *et al.*, 2015). Furthermore, food products may be contaminated through a dirty home environment, contaminated fabric carpets, contact with currency while handling food, via skin surface, dust, aerosols, and through person-to-person transmission (Todd *et al.*, 2009).

Overall, beef sausage recorded the highest prevalence of *E. coli* contamination (Household: 60%; Retail: 50%), bacon recorded the highest prevalence of *L. monocytogenes* (household: 50%; retail: 40%). In contrast, the prevalence of *E. coli* and *L. monocytogenes* remains the same in salami (household: 30%; retail: 20%). The level of *Salmonella* spp. was low in all the processed meat categories and locations.

Bacteria are frequently associated with ham,

Table 1. Microbiological quality (log CFU/g) of the 60 processed meat products of household and retail samples.

Microorganism	Household			Retail		
	Beef sausage	Salami	Bacon	Beef sausage	Salami	Bacon
S. aureus	2.29 ± 0.25^{a}	1.90 ± 0.44^{a}	2.25 ± 0.25^{a}	2.08 ± 0.45^{a}	0.62 ± 0.32^{b}	1.12 ± 0.46^{b}
Anaerobic spore-formers	1.39 ± 0.23^{a}	1.18 ± 0.27^{a}	1.47 ± 0.28^{a}	1.32 ± 0.14^{a}	1.40 ± 0.99^{a}	1.49 ± 0.13^{a}
Aerobic spore-formers	1.54 ± 0.17^{a}	1.59 ± 0.13^{a}	1.72 ± 0.15^{a}	1.17 ± 0.92^{a}	1.24 ± 0.17^{a}	1.22 ± 0.11^{a}

Results are presented as mean ± standard deviation. Same superscript letters in rows are not significantly different ($p > 0.05$).

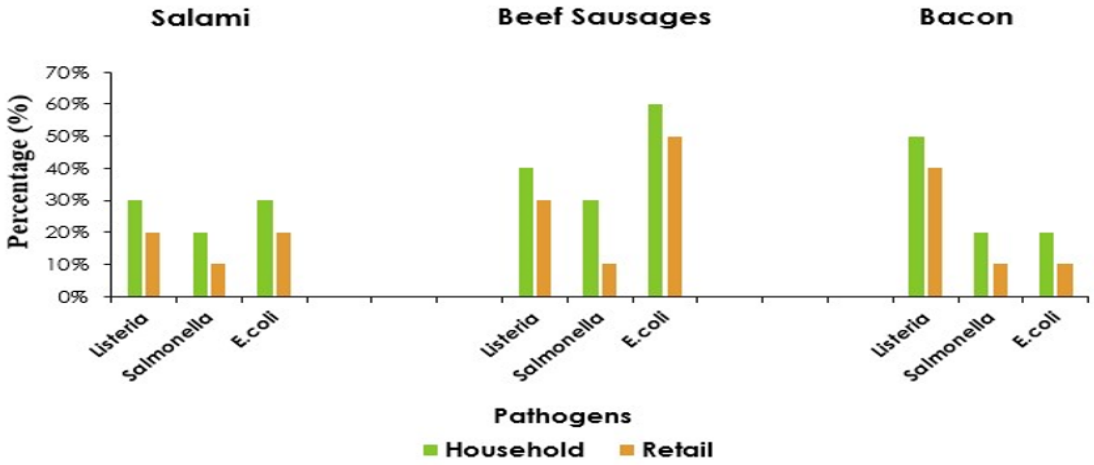

Figure 1. Prevalence and distribution of food pathogens (*L. monocytogenes, Salmonella* spp. and *E. coli*) in RTE meat samples collected in Durban metropolis.

corned beef, salami, bacon, and barbecued meats (Stefani *et al.*, 2012; Ge *et al.*, 2017) because these products are excellent substrates which provide adequate nutrients for the growth of microorganisms leading to food infections and diseases.

Salmonella spp., *L. monocytogenes*, and *E. coli* O157:H7 are the main foodborne pathogens with the highest incidence worldwide, resulting in human illnesses by ingestion of contaminated food. Consequently, about 40% of the approximately 50 million annual deaths, mostly in developing countries, occur due to consumption of contaminated food (Zaulet *et al.*, 2016). Furthermore, viable cells, even if present in small numbers, have the potential to multiply in the digestive tract and cause severe gastroenteritis (Igumbor *et al.*, 2012). The presence of these pathogens is an indicator of poor sanitation and suggests that retail outlets and households are not maintaining the desired level of hygienic conditions and may be unaware of the implications of unhygienic practices (Almanza *et al.*, 2007). In addition, training of the staff on food hygienic procedures at a food processing facility is required to eliminate cross-contamination at the manufacturing and retail levels (Mani-López *et al.*, 2012). Cross-contamination between meat and surfaces is a multifactorial process and strongly depends on the species, initial contamination level, types of surface, contact time, and the number of the subsequent fillets. Thus, quantifying the cross-contamination risk associated with various steps of meat processing and food establishments or households can provide a scientific basis for risk management of such products (Gkana *et al.*, 2017). The *16s rRNA* sequencing confirmed the presence of *L. monocytogenes*.

Food microbial contamination is now recognised as a major public health threat all over the world. A food handling habit and contaminated environment affect food products during processing and storage. This can result in the new transmission-contamination cycles as proper sanitation is not accessible by half of the rural population (South African Government, 2004).

According to Leong *et al.* (2016), an amendment in the South African Foodstuffs, Cosmetics, and Disinfectants Act (South African Government, 1972) speaks nothing on the microbiological standards of *Listeria* spp. but South African voluntary standard, South African National Standard (SANS, 2011) allows the maximum limit of 100 CFU/g at the end of shelf-life of *L. monocytogenes* in processed meat products. Similarly, the South African microbial guidelines do not make provision for maximum counts related to *E. coli* on food contact surfaces or hands but have recommended that this pathogen must be absent in all food products (Lambrechts *et al.*, 2014). Many of the important features of good food safety management, including the maintenance of a positive food safety culture, have not yet been adopted in South Africa. Although an official system of internal hygiene auditing exists and food safety training is provided to food handlers, they have not yet been integrated into a comprehensive approach for food safety management (Griffith *et al.*, 2017). According to WHO (2017), processed meat products are susceptible to an array of microbial hazards. Although these hazards may not be significant due to extreme thermal processing conditions of meat, many pathogens can still survive and remain viable for extended periods of time, posing a risk to consumers (Abdul-Raouf *et al.*, 1993). The microorganisms identified in meat products via *16s rRNA* sequencing included *Enterobacter cloacae* (MK685145), *Lelliottia amnigena* (MK685141),

Table 2. The effect of natural antimicrobials on *E. coli* O157:H7 and *L. monocytogenes* in a broth culture over a period of 72 h at 4°C and 30°C.

Microorganism /Natural antimicrobial	Storage temperature (4°C)				Storage temperature (30°C)			
	Population (log CFU/g) over the period of storage (h)				Population (log CFU/g) over the period of storage (h)			
	0	24	48	72	0	24	48	72
E. coli								
Lemon juice	8.37 ± 0.08^{a}	8.34 ± 0.03^{b}	7.98 ± 0.03^{b}	7.20 ± 0.03^{b}	9.37 ± 0.08^{a}	8.37 ± 0.11^{b}	8.28 ± 0.11^{b}	8.25 ± 0.11^{b}
Vinegar	9.38 ± 0.01^{a}	8.29 ± 0.05^{b}	7.87 ± 0.05^{b}	7.22 ± 0.05^{b}	9.38 ± 0.01^{a}	8.40 ± 0.01^{b}	8.33 ± 0.08^{b}	8.27 ± 0.01^{b}
Lemon and vinegar (1:1)	9.36 ± 0.03^{a}	8.18 ± 0.07^{b}	7.14 ± 0.06^{c}	7.01 ± 0.06^{b}	9.36 ± 0.03^{a}	8.34 ± 0.04^{b}	8.11 ± 0.05^{b}	8.09 ± 0.04^{b}
Control	9.44 ± 0.34^{a}	9.50 ± 0.03^{b}	9.53 ± 0.03^{a}	9.58 ± 0.02^{a}	9.44 ± 0.34^{a}	9.44 ± 0.03^{a}	9.45 ± 0.06^{a}	9.46 ± 0.02^{a}
L. monocytogenes								
Lemon juice	9.37 ± 0.08^{a}	8.40 ± 0.02^{a}	7.92 ± 0.02^{b}	7.23 ± 0.02^{b}	9.37 ± 0.08^{a}	8.42 ± 0.02^{b}	8.33 ± 0.10^{b}	8.35 ± 0.10^{b}
Vinegar	9.38 ± 0.01^{a}	8.37 ± 0.08^{a}	7.73 ± 0.08^{b}	7.25 ± 0.08^{b}	9.38 ± 0.01^{a}	8.44 ± 0.07^{b}	8.35 ± 0.06^{b}	8.31 ± 0.06^{b}
Lemon and vinegar (1:1)	9.36 ± 0.03^{a}	8.21 ± 0.05^{a}	7.18 ± 0.05^{c}	7.08 ± 0.08^{b}	9.36 ± 0.03^{a}	8.39 ± 0.09^{b}	8.15 ± 0.08^{b}	8.18 ± 0.08^{b}
Control	9.44 ± 0.34^{a}	9.50 ± 0.03^{a}	9.53 ± 0.03^{a}	9.58 ± 0.02^{a}	9.44 ± 0.34^{a}	9.46 ± 0.01^{a}	9.45 ± 0.03^{a}	9.46 ± 0.02^{a}

Results are presented as mean ± standard deviation. Means within the same column followed by the same letters are not significantly different ($p > 0.05$).

Citrobacter freundii (MK685155), *L. monocytogenes* (MK685142, MK685215), and *Hafnia alvei* (MK685123). The presence of these microorganisms signifies contamination from soil, polluted water, and handlers; thus regulation should be in place to reduce the incident of these contaminants.

Effect of treatments on the survival of E. coli O157:H7 ATCC 43888 and L. monocytogenes ATCC 7644 in a broth system

Table 2 shows the effect of treatments on *E. coli* O157:H7 and *L. monocytogenes* in a broth culture incubated at two different temperatures (4°C and 30°C) over a period of 72 h. *E. coli* O157:H7 demonstrated a slightly higher log reduction than *L. monocytogenes* during the incubation period. At the end of 72 h, there was no significant difference between the lemon juice and vinegar treatments, but the combined treatment had a higher log-reduction of both pathogens with a 2-log reduction at 4°C and 1-log reduction at 30°C. The differences in the log reduction may be due to the effects of higher temperatures leading to the reduced activity and the increased metabolic activity of the pathogen at 30°C.

Although statistically non-significant ($p > 0.05$), vinegar showed higher log-reduction than lemon juice. A similar observation was made by Li *et al.* (2012), who observed a higher antimicrobial efficacy of vinegar than lemon juice. In some studies, the use of vinegar was found to be more effective than lemon juice for inhibiting *E. coli* O157:H7 (Mani-López *et al.*, 2012). This could be due to the differences in variables including type of acidulant, concentration, pH, temperature, and contact time (Yang *et al.*, 2009). Gram-negative bacteria, such as *E. coli* are more susceptible to antimicrobial agents than Gram-positive bacteria, probably due to a thinner cell wall (Chen *et al.*, 2017). However, the synergetic effect of lemon and vinegar may contribute to higher log reduction. The rapid growth of the pathogens observed at 30°C may be attributed to the growth temperature that can easily encourage any mesophilic microorganism to thrive. Almanza *et al.* (2007) reported that bacteria grow rapidly in the range of temperature between 30°C and 40°C, doubling in number in as little as 20 min. In a separate study conducted by Carroll *et al.* (2007), similar results were observed regarding the effect of nisin and lysozyme on foodborne pathogens at storage temperatures of 7°C and 30°C.

Effect of treatments on the survival of E. coli O157:H7 ATCC 43888 and L. monocytogenes ATCC 7644 in salami

Tables 3 and 4 show the effect of lemon juice and vinegar on *E. coli* O157:H7 and *L. monocytogenes* on the surface inoculated salami that was stored at two temperatures (4°C and 7°C) for a period of 144 h. After 144 h, the antimicrobial demonstrated about 5-log reduction at 4°C and 2-log reduction at 7°C. However, there was no significant difference between the individual use of lemon and vinegar but there was a significant difference with the combined use. The synergetic effect of the combined antimicrobials against these foodborne pathogens indicates a

Table 3. The effect of natural antimicrobials on salami inoculated with *E. coli* O157:H7 and *L. monocytogenes* over a period of 144 h at 4°C.

Type of microorganism / Natural antimicrobial	Population (log CFU/g) over a period of storage (h)						
	0	24	48	72	96	120	144
E. coli							
Lemon juice	9.37 ± 0.019^{a}	9.26 ± 0.003^{b}	8.23 ± 0.036^{b}	7.20 ± 0.036^{b}	6.18 ± 0.040^{b}	5.13 ± 0.013^{b}	4.21 ± 0.073^{b}
Vinegar	9.37 ± 0.004^{a}	9.26 ± 0.100^{b}	8.24 ± 0.033^{b}	7.22 ± 0.033^{b}	6.20 ± 0.019^{b}	5.16 ± 0.063^{b}	4.19 ± 0.113^{b}
Lemon and vinegar (1:1)	9.33 ± 0.021^{a}	9.25 ± 0.053^{b}	8.22 ± 0.018^{b}	7.02 ± 0.018^{b}	5.89 ± 0.043^{c}	5.00 ± 0.047^{c}	4.01 ± 0.074^{c}
Control	9.39 ± 0.043^{a}	9.39 ± 0.016^{a}	9.37 ± 0.016^{a}	9.38 ± 0.016^{a}	9.37 ± 0.020^{a}	9.39 ± 0.020^{a}	9.45 ± 0.044^{a}
L. monocytogenes							
Lemon juice	9.37 ± 0.019^{a}	9.29 ± 0.009^{a}	8.28 ± 0.161^{b}	7.38 ± 0.016^{b}	6.31 ± 0.035^{b}	5.27 ± 0.015^{b}	4.47 ± 0.020^{b}
vinegar	9.37 ± 0.004^{a}	9.27 ± 0.007^{a}	8.29 ± 0.158^{b}	7.36 ± 0.015^{b}	6.29 ± 0.017^{b}	5.22 ± 0.013^{b}	4.38 ± 0.109^{b}
Lemon and vinegar (1:1)	9.33 ± 0.021^{a}	9.27 ± 0.004^{a}	8.18 ± 0.016^{b}	7.11 ± 0.016^{c}	6.00 ± 0.036^{c}	5.01 ± 0.017^{c}	4.12 ± 0.062^{c}
Control	9.39 ± 0.043^{a}	9.39 ± 0.016^{a}	9.37 ± 0.016^{a}	9.38 ± 0.016^{a}	9.37 ± 0.020^{a}	9.39 ± 0.020^{a}	9.45 ± 0.044^{a}

Results are presented as mean ± standard deviation. Means with the same column followed by the same superscript letters are not significantly different ($p > 0.05$).

Table 4. The effect of natural antimicrobials on salami inoculated with *E. coli* O157:H7 and *L. monocytogenes* over a period of 144 h at 7°C.

Type of microorganism / Natural antimicrobial	Population (log CFU/g) over a period of storage (h)						
	0	24	48	72	96	120	144
E. coli							
Lemon juice	9.35 ± 0.005^{a}	9.26 ± 0.005^{a}	8.25 ± 0.034^{b}	8.22 ± 0.034^{c}	7.19 ± 0.039^{c}	7.17 ± 0.083^{c}	7.14 ± 0.089^{c}
vinegar	9.32 ± 0.032^{b}	9.29 ± 0.032^{b}	8.27 ± 0.033^{a}	8.25 ± 0.035^{b}	7.23 ± 0.036^{b}	7.19 ± 0.079^{b}	7.17 ± 0.041^{b}
Lemon and vinegar (1:1)	9.33 ± 0.001^{b}	9.25 ± 0.001^{b}	8.24 ± 0.053^{b}	8.21 ± 0.053^{c}	7.18 ± 0.060^{d}	7.15 ± 0.043^{d}	7.20 ± 0.070^{d}
Control	9.40 ± 0.021^{b}	9.39 ± 0.016^{b}	9.43 ± 0.016^{a}	9.62 ± 0.016^{a}	9.79 ± 0.020^{a}	9.89 ± 0.020^{a}	9.94 ± 0.044^{a}
L. monocytogenes							
Lemon juice	9.35 ± 0.005^{a}	9.30 ± 0.001^{b}	8.28 ± 0.048^{b}	8.26 ± 0.004^{b}	7.25 ± 0.034^{c}	7.23 ± 0.054^{c}	7.18 ± 0.040^{c}
vinegar	9.32 ± 0.032^{b}	9.31 ± 0.029^{a}	8.30 ± 0.015^{a}	8.28 ± 0.015^{a}	7.26 ± 0.030^{b}	7.25 ± 0.034^{b}	7.19 ± 0.077^{b}
Lemon and vinegar (1:1)	9.33 ± 0.001^{b}	9.30 ± 0.015^{d}	8.29 ± 0.030^{b}	8.27 ± 0.063^{b}	7.24 ± 0.071^{d}	7.23 ± 0.018^{d}	7.16 ± 0.085^{d}
Control	9.40 ± 0.021^{b}	9.39 ± 0.016^{b}	9.43 ± 0.016^{a}	9.62 ± 0.016^{a}	9.79 ± 0.020^{a}	9.89 ± 0.020^{a}	9.94 ± 0.044^{a}

Results are presented as mean ± standard deviation. Means with the same column followed by the same superscript letters are not significantly different ($p > 0.05$).

bactericidal activity at 4°C and bacteriostatic activity at 7°C. Furthermore, the cold storage temperature can influence the antimicrobial efficiency and affect the quality of a product.

The higher log counts of *E. coli* O157:H7 and *L. monocytogenes* in salami at 7°C as compared to 4°C are in agreement with the studies of Almanza *et al.* (2007), who experienced higher growths at 7°C storage temperature than 4°C storage temperature of the samples.

Studies suggest that the exposure of foodborne pathogens to an irregular storage environment may result in survivors and increased tolerance to acidic conditions in food substrate (Mani-López *et al.*, 2012). Although these pathogens grow poorly at 4°C, an increase in growth rate is encouraged at temperatures < 4°C (O'Connell *et al.*, 2016).

Conclusion

Staphylococcus aureus, aerobic, and anaerobic spore formers were identified in both household and retail samples but the prevalence of *S. aureus* was lower in the retail samples (salami and bacon)

than the household samples. Beef sausage showed the highest prevalence for all pathogens tested. However, *L. monocytogenes* prevalence was higher in household samples while *E. coli* was observed to be higher in the retail samples signifying that both retail and household samples present a public health risk. The higher log reduction in broth culture at 4°C suggests that the antimicrobials may not be very effective at high temperature (30°C). Temperature, contact time, and combinations of different antimicrobials should be considered when applying treatments for RTE foods. The analysis of the antimicrobial activity against the test pathogens on salami indicated better antimicrobial effect at 4°C than 7°C. Therefore, temperature abuse must be avoided to achieve an effective antimicrobial effect. Furthermore, the higher log reduction observed with the combined use of vinegar and lemon suggests the efficacy of a synergetic effect of these antimicrobials. There should be regular and effective sensitisation for handling and storage of RTE foods, especially at the household level.

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