Article

Bulk sales of cold cuts and sausages: a marketing trend associated to the risk of foodborne diseases in Culiacan, Mexico

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Abstract:

The nature, production and consumption of cold cuts and sausages define these foods as vulnerable to contamination by pathogenic microorganisms that cause foodborne diseases. The aim of the study was to evaluate the influence of the bulk sale of cold cuts and sausages, on the hygienic quality and prevalence of *L. monocytogenes* and *Salmonella*. Thus, 96 samples of sausages (sausage and ham) from 15 national brands were collected and classified by type of sale: original package (n= 48) and bulk (n= 48). The detection of total coliforms, *Escherichia coli, Salmonella*, and *L. monocytogenes* was analyzed by traditional culture method. 42.7 % (41/96) of the samples of cold cuts and sausages failed to comply with the sanitary specifications in terms of total coliforms and *E. coli*. The statistical analysis showed that the type of sale is an indicator of the microbial risk in cold cuts and sausages ($\chi 2 = 40.0$, *P*= 0.000), since the number of samples with poor hygienic quality was higher for bulk sales (69.0%) compared to those sold in packages (17.0%). Additionally, the risk analysis showed that bulk sale increases 41.8 and 5.9 times the risk of acquiring ham and frankfurters of poor

microbiological quality (P < 0.05), respectively, while the type of sales did not influence the presence of *L. monocytogenes* (6.3 %). Consumers are advised to avoid bulk sale of cold cuts and sausages, and producers and sellers should reinforce good hygienic practices that will ensure food safety, and minimize the risk of infection.

Key words: Bulk, L. monocytogenes, Package, Food safety, Cold cuts, Cold sausages.

Received: 21/02/2019

Accepted: 29/08/2019

Introduction

Ready-to-eat (RTE) cold cuts and sausages are foods of great acceptance due to their organoleptic attributes and practicality of consumption. In Mexico, the *per capita* consumption of these products is increasing, since it is estimated that in 2014, 6.9 kg per person were consumed, while for 2018 the figure increased to 8.1 kg⁽¹⁾. However, the nature and the production of cold cuts and sausages render these foods vulnerable to contamination by microorganisms that cause their deterioration, as well as by pathogens associated to foodborne diseases (FBD)⁽²⁾. *Listeria monocytogenes* is considered the main cause of withdrawal of cold cuts and sausages from the market in the USA.⁽³⁻⁶⁾, and one of the main etiological agents of FBD, with a mortality rate of up to 30 % in certain high-risk groups (immunocompromised, elderly, infants, and pregnant women)^(2,4-6). It has been reported as the cause of 1,455 cases (16 % mortality)⁽⁷⁾, with an estimated economic cost of US \$ 2.4 billion due to annual listeriosis⁽⁸⁾. However, there are no official references for such estimates in Mexico.

Fortunately, the Mexican Official Norm Project PROY-NOM-213-SSA1-2017⁽⁹⁾, which already establishes the monitoring of this pathogen, has been published. However, because this project is recent, there is little information on the prevalence of *Listeria monocytogenes* in cold cuts and sausages^(10,11). Furthermore, the National Epidemiological Surveillance System does not consider listeriosis as a mandatory notification disease, making it difficult to establish relationships between the presence of the bacteria in food and cases of disease. Even so, the microbiological quality of ham and sausage has already been questioned by the presence of aerobic mesophilic organisms, as well as of pathogenic microorganisms (*Salmonella, Staphylococcus aureus*)⁽¹²⁾. Despite this, there has been no research aimed at establishing the origin of the microbiological contamination of these foods.

In order to guarantee the safety of cold cuts and sausages, the production process adheres to good hygienic practices and to the analysis of dangers and critical control points⁽⁴⁾. In spite of this, poor hygienic handling of these foods in retail stores has been associated as the cause of most reported epidemiological outbreaks⁽⁵⁾.

Several brands of cold cuts and sausages are available in Mexico, which can be sold in bulk or in their original package. In particular, the Mexican consumers' habit of acquiring cold cuts and sausages in bulk is a worrying aspect, given that it involves handling the food at the point of sale, and violating the brand's quality guarantee. Consequently, the population most affected is the one that seeks to save resources through this practice, and, in addition, does not go to see a doctor in case of contracting an FBD. Therefore, the aim of this study was to generate information on the influence of the type of sale on health quality (*Salmonella, Escherichia coli*, coliforms)^(9,13) and the prevalence of *L. monocytogenes* in cold cuts and sausages traded in the state of Sinaloa, Mexico.

Material and methods

Samples collection

An intentional sampling was carried out in order to assess the influence of two sales strategies (bulk and package) on cold cuts and sausages. The samples were selected according to the main brands of national distribution and the most popular among consumers. The selected points of sale (markets and supermarkets) were conditioned by the availability of the selected brands, as well as by offering the two types of sale of the product. A total of 96 samples of turkey cold cuts and sausages consisting of sausages (n=48) and ham (n=48) were collected from 15 different national brands (AP) and from 13 points of sale in Culiacan, Mexico, during the second half of 2017, samples of cold cuts and sausages were collected and classified equally by type of sale; original package (n = 48) and bulk (n = 48). The purchase of the "original package" samples was defined as the acquisition of the complete and closed unit of the product. Whereas, the purchase in "bulk" involved the manual handling of the sausage by the seller, the slicing or mechanical cutting, and the subsequent placement of the product in plastic bags provided by the retail service. The food storage temperature was observed to be 5 ± 2 °C, according to the digital readers of the refrigeration equipment at the points of sale. The samples were stored in a refrigerated box during transportation to the laboratory for microbiological analysis during a period of <4 h after collection. The outer surface of the original packaging and the plastic bags provided during the bulk sale were disinfected with alcohol (70%) prior to microbiological analysis.

Quantification of Escherichia coli and total coliforms

Quantification of *E. coli* and total coliforms was performed on 3MTM PetrifilmTM plates according to the supplier's instructions. The quantification of each microorganism was performed according to the typical colonial morphology observed. Microbial concentrations were expressed as CFU/g, and were compared with the microbiological limit reported in NOM-213-SSA1-2002⁽¹³⁾.

Isolation of Salmonella

For the isolation of *Salmonella* in the samples of cold cuts and sausages, the protocol described in the Bacteriological Analytical Manual⁽¹⁴⁾ was followed. The *Salmonella* isolates were confirmed by amplifying a 244 bp fragment of the invA gene by PCR⁽¹⁵⁾.

Isolation of L. monocytogenes

L. monocytogenes was isolated by the protocol described by the USDA-FSIS⁽¹⁶⁾. Presumptive *Listeria* isolates were confirmed with the Microgen Listeria-ID system (Microgen LABTM). Further confirmation of *L. monocytogenes* isolates was performed using the polymerase chain reaction (PCR) method by amplifying a 234 bp fragment of the Listeriolysin-O gene⁽¹⁷⁾. *L. monocytogenes* ATCC 7644 was included as a positive control.

Statistical analysis

IBM® SPSS software was used for the statistical analysis of the results. A non-parametric Pearson $\chi 2$ test was performed to determine the association of the type of sale with the microbiological quality of the sausage. The risk analysis was performed by calculating the odds ratio. A value of *P* <0.05 was considered statistically significant.

Results

Table 1 shows the microbiological quality of the samples of cold cuts and sausages according to the type of sale and sausage. In general, 42.7 % (41/96) of the sausage samples included in the bulk (69.0 %) and package (17.0 %) category were outside the sanitary specification (<3 MPN/g) (9.13), since the values of quantification for *E. coli* and total coliforms were in the range of 10 to 2,860 CFU/g. The incidence rate of samples of sausage and ham that did not meet the health specification ranged from 29.2 % (14/38) to 56.3 % (27/48), respectively. *Salmonella* was not detected in any of the samples analyzed. Additionally, 6.3 % (6/96) of the samples were positive for *L. monocytogenes*; bulk (8.3 %) *vs* package (4.2 %). It should be noted that 60.0% (9/15) of the national brands are outside the sanitary specification^(9,13), and four of these brands (26.6 %) were positive for *L. monocytogenes*.

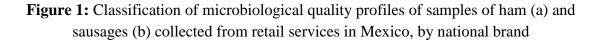
1n bulk in Mexico									
	Detection (%) of micro	organisms						
Microbiological	Package		Bulk		Tatal				
criterion *	Sausage	Ham	Sausage	Ham	— Total (n=96)				
	(n=24)	(n=24)	(n=24)	(n=24)	(11=90)				
Coliforms	8.3	4.2	45.8	91.7	37.5				
E. coli	4.2	0	4.2	12.5	5.2				
Salmonella	0	0	0	0	0				
L. monocytogenes	4.2	4.2	4.2	12.5	6.3				
Rejected**	12.0	21.0	46.0	92.0	42.7				
Accepted	88.0	79.0	54.0	8.0	57.3				

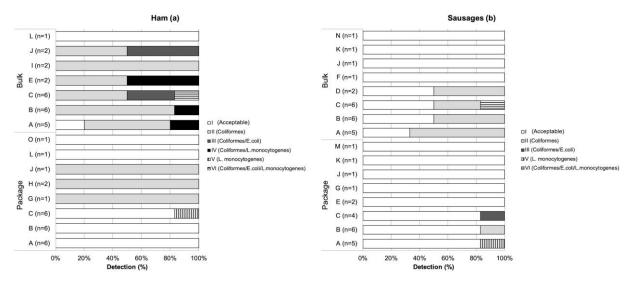
Table 1. Microbiological quality of samples of cold cuts and sausages sold in package and in bulk in Mexico

*The presence of coliforms and *E. coli* was based on the sanitary limit (<3 NMP/g for fecal coliforms) for cooked foods defined by NOM-213-SSA1-2002.

**The samples classified as rejected contained at least one microorganism.

The microbiological quality profiles highlight that the type of sale is an indicator of the assurance of the safety of most brands of cold cuts and sausages ($\chi 2 = 40.0$, P = 0.000) (Figure 1) in terms of coliforms, the bulk sale being a microbiological risk factor, mainly in ham. The type of sale did not influence the presence of *L. monocytogenes* and *E. coli* (*P*<0.05) in the samples of cold cuts and sausages. The risk analysis (odd ratio) determined that the sale in bulk increases 11 times the risk of acquiring a non-innocuous sausage, compared to the sale of the product in its original package. Likewise, the risk analysis by type of sausage shows that selling in bulk increases 41.8 and 5.9 times the risk of acquiring ham and sausages of poor microbiological quality, respectively (Table 2). No significance was observed between the risk of obtaining bulk or package products contaminated with *L. monocytogenes*; however, this risk may increase by 1-3 times if bulk purchase is chosen.





The letters A-O refer to national brands, and the number of samples per brand is in parentheses. The variability of the number of samples per category depended on the availability of the sample.

Category	Total coliforms a	nd <i>E. coli</i>	L. monocytogenes	
	Odd ratio	Significance	Odd ratio	Significance
	(IC 95%)	Р	(IC 95%)	Р
Cold cuts	11.0	0.0001	2.1	0.4079
	(4.15 - 29.13)		(0.36 – 11.99)	
Ham	41.8	0.0001	3.3	0.9970
	(7.26 - 240.78)		(0.32 – 34.08)	
Sausage	5.9	0.0163	1.0	1.0000
	(1.39 – 25.30)		(0.06 – 16.97)	

Table 2: Estimation of the risk of acquiring non-innocuous cold cuts and sausages for sale in bulk in Mexico

Discussion

The microbiological quality profiles revealed that most of the brands sold in Culiacan, Sinaloa did not meet the sanitary specifications required for their consumption⁽⁹⁾. This indicates the poor hygienic conditions in which some cold cuts and sausages are processed (marks A, B, C, H), and the influence of the integrity of the original packaging during the sale on the microbiological quality. Although primary contamination of cold cuts and

sausages may $occur^{(18)}$, their subsequent handling in establishments and at home can increase the pathogenic microbial $load^{(4-5)}$.

In this regard, the violation of the original packaging, the manual handling by the seller, the slicing and cutting, and the use of non-sanitary packaging for the bulk sale of cold cuts and sausages are attributes observed in the study and are pointed out as factors of microbial contamination of food⁽⁵⁾. The detection of *L. monocytogenes* and the count of coliforms and *E. coli* in cold cuts and sausages are indicators of the risk of the development of listeriosis and gastroenteritis, since these foods can be consumed without heat treatment prior to their intake. It is important to highlight that these brands are distributed nationally, and that the habit of buying in bulk is a common practice carried out by the Mexican population, which exhibits a potential public health problem.

An equal risk of contamination with *L. monocytogenes* has been observed regardless of whether the cold cuts and sausages are acquired in bulk or in packaging; this defines the pathogen as a potential contamination hazard at any stage of the food chain. Kurpas *et al*⁽⁴⁾ agree that the final contamination of meat products by *L. monocytogenes* occurs at both the processing plant and the point of sale. This is due to the growth characteristics, propagation pathways and reservoirs of *L.* monocytogenes, as well as to the poor hygiene of the handler, since this bacterium is persistent in conditions related to food processing⁽⁴⁾. Food contamination with L. *monocytogenes* is one of the most important challenges faced by the meat industry, due to the bacterium's ability to multiply during refrigerated food storage, and to the fact that a contaminated food may be consumed without additional cooking⁽¹⁹⁾. Many studies have reported the presence of *L. monocytogenes* in cooked, raw, cured, salted cold cuts and sausages, among others^{(18,20).}

Other food safety concerns with regard to *L. monocytogenes* are virulence⁽²¹⁾ and antibiotic resistance⁽²²⁾, both of which are important factors in the severity of listeriosis and the persistence of the pathogen in the processing environment. These phenotypic attributes have also been previously reported in *L. monocytogenes* strains recovered from certain foods in $Mexico^{(10,11)}$.

Attention should be paid to the detection of *L. monocytogenes* in cold cuts and sausages sold in Mexico, since the clinical and epidemiological importance of listeriosis in the Mexican population has been previously documented, highlighting the high mortality rate and the severity of clinical manifestations⁽²³⁾. Listeriosis can be present in the form of sepsis, meningitis, endocarditis, miscarriage, localized infections and gastroenteritis⁽²⁴⁾. Additionally, Castañeda *et al*⁽²⁵⁾ have established the clonal relationship of a strain of *L. monocytogenes* isolated from chicken meat as a potential etiological agent of listeriosis in Mexico. Since *L. monocytogenes* is considered a pathogen responsible for ATS⁽⁷⁾, the prevention and intentional control of the pathogen in the food chain is a guideline for minimizing infection by this bacterium.

Since the sale in bulk is an indicator of high microbiological risk of cold cuts and sausages sold in retail stores, attention must be paid to this habit of buying by Mexican consumers, and actions should be designed to help minimize the risk of infection. The high risk of acquiring non-innocuous cold cuts or sausages due to purchase in bulk denotes the continuous exposure of consumers to microbiological hazards, and proposes this fact as a potential cause of the cases of intestinal infections (5,771,681) and food poisoning (38,815) reported annually in Mexico, and whose classification is not defined⁽²⁶⁾. The fulfillment of the regulatory provisions in the sale of these products, and the application of the protocols of good hygienic practices guarantee the safe sale to the consumers^(4,9,13). Some studies suggest that sausages must be sold in their original packaging to maintain their production guarantee⁽⁴⁾. Additionally, consumer education on safety issues is an important action for teaching the proper handling of food and minimizing the risk of FBD⁽²⁷⁾.

Conclusions and implications

The high level of microbial contamination and the detection of *L. monocytogenes* in cold cuts and sausages of different national brands commercialized in bulk or package, is a common indicator of the lack of good hygiene practices in the food chain, and a high risk of acquiring FBD, since they are ready-to-eat products and are distributed throughout the country. For this reason, it is imperative to call the attention of the Mexican authorities to demand the monitoring of *L. monocytogenes* in cold cuts and sausages, as well as to implement microbial control strategies in the formulation and sale. Finally, consumers are encouraged to choose products in their original packaging to maintain food safety and brand assurance.

Acknowledgements

This work was supported by a grant from the PROFAPI2014 / 043 Research Projects Promotion and Support Program of the Autonomous University of Sinaloa (Universidad Autónoma de Sinaloa).

Conflict of interests

The authors declare that they have no conflicts of interest, financial or otherwise.

Literature cited:

- 1. COMECARNE. Consejo Mexicano de Carne. Compendio Estadístico 2018. México. 2018. https://comecarne.org/estadisticas/. Consultado 19 Sep, 2018.
- 2. Olaoye OA. Meat: An overview of its composition, biochemical changes and associated microbial agents. Int Food Res J 2011;18(3):877-885.
- USDA-FSIS. United States Department of Agriculture Food Safety and Inspection Service. Recall Case Archive. http://www.fsis.usda.gov/wps/portal/fsis/topics/recallsand-public-health-alerts/recall-case-archive. Accessed Sep 19, 2018.
- 4. Kurpas M, Wieczorek K, Osek J. Ready-to-eat meat products as a source of *Listeria monocytogenes*. J Vet Res 2018;61(1):49-55.
- Endrikat S, Gallagher D, Pouillot R, Quesenberry HH, Labarre D, Schroeder CM, Kause J. A Comparative risk assessment for *Listeria monocytogenes* in prepackaged *versus* retail-sliced deli meat. J Food Prot 2010;73(4):612–619.
- Leonge D, Alvarez-Ordoñez A, Jooste P, Jordan K. *Listeria monocytogenes* in food: Control by monitoring the food processing environment. Afr J Microbiol Res 2016;10:101–114.
- 7. Scallan E, Griffin P, Angulo F, Tauxe R, Widdowson M, Roy S, Jones J, Griffin P. Foodborne illness acquired in the United States. Emer Infect Dis 2011;17:7–15.
- 8. Ivanek R, Grohn YT, Tauer LW. The cost and benefit of *Listeria monocytogenes* food safety measures. Crit Rev Food Sci Nutr 2004;44:513–523.
- SSA. Secretaría de Salud Proyecto de Norma Oficial Mexicana, Productos y servicios. Productos cárnicos procesados y los establecimientos dedicados a su proceso. Disposiciones y especificaciones sanitarias. Métodos de prueba. PROY-NOM-213-SSA1-2017. México. 2017.
- Castañeda GM, Castro N, León J, Valdez JB, Guzmán JR, Luchansky JB, Porto-Fett ACS, Shoyer AB, Chaidez C. Prevalence, levels, and relatedness of *Listeria monocytogenes* isolated from raw and ready-to-eat foods at retail markets in Culiacan, Sinaloa, México. J Microbiol Res 2013;3(2):92–98.

- 11. Silva LE, Pérez C, Barreras A, Figueroa F. Identification of *Listeria sp.* in hams and frankfurters products exhibited for sale. J Anim Vet Adv 2007;6(3):314–316.
- 12. Sarti E, Parrilla MC, Saldate O. Calidad sanitaria del jamón que se consume en la ciudad de México. Salud Públ Méx 1989;31(3):326-333.
- 13. SSA. Secretaría de Salud. Productos y Servicios. Productos cárnicos procesados. Especificaciones sanitarias. Métodos de prueba. NOM-213-SSA1-2002. México. 2005.
- BAM. Bacteriological Analytical Manual. Chapter 5: Salmonella. Bacteriological Analytical Method. USA. 2018. https://www.fda.gov/food/laboratory-methodsfood/bacteriological-analytical-manual-bam-chapter-5-salmonella. Accessed Sep 19, 2017.
- 15. Chiu C, Ou J. Rapid identification of *Salmonella* serovars in feces by specific detection of virulence genes *invA* and *spvC*, by an enrichment broth culture-multiplex PCR combination assay. J Clin Microbiol 1996;34(10):2619–2622.
- 16. United States Department of Agriculture-Food Safety and Inspection Services (USDA-FSIS). 2017. MLG8: Isolation and identification of *Listeria monocytogenes* from red meat, poultry, egg and environmental samples. http://www.fsis.usda.gov/wps/wcm/connect/1710bee8-76b9-4e6c-92fc-fdc290dbfa92/MLG-8.pdf?MOD=AJPERES_ Accessed Sep 19, 2017.
- 17. Furrer B, Candrian U, Hoefelein C, Luethy J. Detection and identification of *Listeria monocytogenes* in cooked sausage products and in milk by in vitro amplification of haemolysin gene fragments. J Appl Bacteriol 1991;70(5):372-379.
- Syne S, Ramsubhag A, Adesiyun A. Microbiological hazard analysis of ready-to-eat meats processed at a food plant in Trinidad, West Indies. Infect Ecol Epidemiol 2013;3(1):1–12.
- 19. Sofos JN. Challenges to meat safety in the 21st century. Meat Sci 2008;78(1-2):3–13.
- 20. Gómez D, Iguácel LP, Rota MC, Carramiñana JJ, Ariño A, Yangüela J. Occurrence of *Listeria monocytogenes* in ready-to-eat meat products and meat processing plants in Spain. Foods 2015;4(3):271–282.
- 21. Khan JA, Rathore RS, Khan S, Ahmad I. *In vitro* detection of pathogenic *Listeria monocytogenes* from food sources by conventional, molecular and cell culture method. Braz J Microbiol 2013;44(3):751–758.
- Lungu BC, O'bryan A, Muthaiyan A, Milillo SR, Johnson MG, Crandall PG, Ricke SC. Listeria monocytogenes: Antibiotic resistance in food production. Foodborne Pathog Dis 2011;8(5):569–578.

- 23. Castañeda-Ruelas G, Eslava C, Castro N, León J, Chaidez C. Listeriosis en México: importancia clínica y epidemiológica. Salud Públ Méx 2014;56(6):654–659.
- 24. Doganay M. Listeriosis: clinical presentation. FEMS Immunol Med Microbiol 2003;35(3):173-175.
- 25. Castañeda-Ruelas G, Eslava C, Castro N, León J, Chaidez C. *Listeria monocytogenes* y Listeriosis, problema de salud pública en México. Salud Públ Méx 2018;60(4):376–377.
- 26. DGE. Dirección General de Epidemiología. Distribución de casos nuevos de enfemedad por fuente de notificación Estados Unidos Mexicanos 2017. http://www.epidemiologia.salud.gob.mx/anuario/2017/morbilidad/nacional/distribucio n_casos_nuevos_enfermedad_fuente_notificacion.pdf. Consultado 29 Ene, 2019.
- Nesbitt A, Thomas MK, Marshall B, Snedeker K, Meleta K, Watson B, Bienefeld M. Baseline for consumer food safety knowledge and behaviour in Canada. Food Control 2014;(38):157-173.