



BRIEF REPORT

Prevalence of *Campylobacter jejuni* and *Campylobacter coli* from broilers at conventional and kosher abattoirs and retail stores

Guillermo F. Guirin^a, Victoria Brusa^{b,*}, Cristian D. Adriani^c, Gerardo A. Leotta^b^a Laboratorio AGGA, Ciudad Autónoma de Buenos Aires, Argentina^b IGEVET - Instituto de Genética Veterinaria "Ing. Fernando N. Dulout" (UNLP-CONICET LA PLATA), Facultad de Ciencias Veterinarias UNLP, Calle 60 y 118 s/n, 1900 La Plata, Argentina^c Departamento de Seguridad Alimentaria, Municipalidad de Berisso, Berisso, Argentina

Received 18 March 2019; accepted 13 July 2019

KEYWORDS

Campylobacter;
Chicken;
Abattoir;
Kosher;
Retail store

Abstract We studied and compared the prevalence of *Campylobacter jejuni* and *Campylobacter coli* in chicken carcasses from conventional and kosher broiler abattoirs and retail stores. The prevalence of thermotolerant *Campylobacter*-positive carcasses was 94.0 (kosher) and 32.0% (conventional) ($p < 0.0001$), while the prevalence of samples contaminated with *C. jejuni*, *C. coli* and simultaneously with both species was 36.0, 2.0 and 56.0% (kosher) and 26.0, 4.0 and 2.0% (conventional) ($p < 0.0001$), respectively. Samples of chicken carcasses ($n = 25$) and food contact surfaces (tables, $n = 25$; knives, $n = 25$) from 25 retails were collected and risk quantification was performed. Retails were categorized as high-risk ($n = 11$), moderate-risk ($n = 11$) and low-risk ($n = 3$). Nineteen (76.0%) carcasses, 20 (80.0%) tables and 18 (72.0%) knives were *Campylobacter*-positive. Retails and abattoirs proved to be sources of carcass contamination with *Campylobacter* spp. Carcasses from kosher abattoirs were mostly contaminated with *Campylobacter* spp., whereas *C. coli* was the most prevalent species isolated from carcasses in retail stores.

© 2019 Asociación Argentina de Microbiología. Published by Elsevier España, S.L.U. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

PALABRAS CLAVE

Campylobacter;
Pollo;
Frigorífico;
Kosher;
Locales de expendio

Prevalencia de *Campylobacter jejuni* y *Campylobacter coli* en carcasas de pollo procedentes de frigoríficos, faena tradicional y kosher, y de locales de expendio

Resumen El objetivo del estudio fue determinar y comparar la prevalencia de *Campylobacter jejuni* y *Campylobacter coli* en carcasas de pollo obtenidas en frigoríficos por faena convencional y kosher, y en locales de expendio. La prevalencia de *Campylobacter* spp. termotolerante fue del

* Corresponding author.

E-mail address: toibrusa@hotmail.com (V. Brusa).<https://doi.org/10.1016/j.ram.2019.07.002>0325-7541/© 2019 Asociación Argentina de Microbiología. Published by Elsevier España, S.L.U. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Please cite this article in press as: Guirin GF, et al. Prevalence of *Campylobacter jejuni* and *Campylobacter coli* from broilers at conventional and kosher abattoirs and retail stores. Rev Argent Microbiol. 2019. <https://doi.org/10.1016/j.ram.2019.07.002>

94,0 (kosher) y del 32,0% (convencional) ($p < 0,0001$). La prevalencia de muestras contaminadas con *C. jejuni*, *C. coli* y con ambas especies fue del 36,0, del 2,0 y del 56,0% (Kosher) y del 26,0, del 4,0 y del 2,0% (convencional) ($p < 0,0001$), respectivamente. Se tomaron muestras de carcasas ($n = 25$) y superficies (tablas, $n = 25$; cuchilla, $n = 25$) en 25 locales. Los locales fueron categorizados como de riesgo alto ($n = 11$), moderado ($n = 11$) y bajo ($n = 3$). Diecinueve (76,0%) carcasas, 20 (80,0%) tablas y 18 (72,0%) cuchillas fueron positivas para *Campylobacter* spp. Frigoríficos y locales fueron fuente de contaminación de carcasas con *Campylobacter* spp. La prevalencia de *Campylobacter* spp. fue mayor en carcasas kosher. *Campylobacter coli* fue la especie más prevalente en carcasas de locales.

© 2019 Asociación Argentina de Microbiología. Publicado por Elsevier España, S.L.U. Este es un artículo Open Access bajo la licencia CC BY-NC-ND (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Consumption of contaminated food with *Campylobacter jejuni* subsp. *jejuni* and *Campylobacter coli* is among the four main causes of diarrheal disease worldwide⁴. Thermotolerant *Campylobacter* species are among the most relevant microbiological hazards associated with poultry meat consumption^{13,15}. Although the prevalence of *Campylobacter* has been studied in conventional broiler abattoirs¹⁵, the impact of kosher slaughter procedures on broiler chicken has not been described yet.

The aims of this study were to: (a) determine and compare the presence of *C. jejuni* and *C. coli* at conventional and kosher broiler abattoirs, and (b) determine the presence of *C. jejuni* and *C. coli* in retail stores.

The study was conducted between November 2011 and April 2013 in broiler abattoirs and retail stores from Buenos Aires province, Argentina. Chicken carcass samples were obtained from two broiler abattoirs ($n = 50$ each) that applied conventional and kosher slaughter processes authorized by the National Service of Agrifood Health and Quality of Argentina. Chicken carcasses were collected and sampled using sterile sponges (Whirl-Pak speci-sponge, Nasco, USA) soaked in 10 ml 3M™ *Campylobacter* enrichment broth (3M, MN, USA) containing 3M™ *Campylobacter* supplement for Tecra™ kits (3M, NSW, Australia), according to Baker et al.², with slight modifications, while all carcasses were sponged. At the same time, 25 retail stores were randomly sampled and risk quantification was performed according to Leotta et al.⁸. The study was conducted in full agreement with the local sanitary authorities. The checklist used for risk quantification included five groups of variables (total value, 100): (1) situation and conditions of the building (10.0), (2) equipment and tools (15.0), (3) handlers (25.0), (4) raw materials and products for sale (20.0), and (5) production flow (30.0). Risk assessment on a 1–100 scale was quantified as high-risk (1–40), moderate-risk (41–70), or low-risk (71–100). Samples were taken from chicken carcasses ($n = 25$) and food contact surfaces (tables, $n = 25$; knives, $n = 25$). One chicken carcass from each retail store was sponged as already described for abattoir carcasses. All the carcasses were provided by conventional abattoirs. In addition, three areas of 20 × 20 cm each from tables and the entire surface of knife blades were sponged.

Each abattoir and retail sponge were incubated with 150 ml *Campylobacter* 3M broth + supplement under microaerophilic conditions (5% O₂, 10% CO₂ and balance N₂) for 24 h at 42 °C. After incubation, DNA was extracted using the Short Prep II kit (Bioteccon, Postdam, Germany). Real-time (RT) PCR was performed using Foodproof® *Campylobacter* 5'Nuclease (Bioteccon) in a thermal cycler (Mx3005P, General Electric, Massachusetts, USA). Positive samples were placed onto Karmali agar (Oxoid, Thermo Scientific, UK) containing *Campylobacter* supplement (Oxoid). Plates were incubated as mentioned above for sponges. All the isolated strains were identified and characterized using the API Campy system (bioMérieux, Marcy l'Etoile, France) and RT-PCR (Bioteccon) as previously described.

The association between *Campylobacter*-positive chicken carcass samples and slaughter process (conventional vs. kosher) was determined using the Chi-square test. The Fisher's exact test was used to evaluate the association of *Campylobacter*-positive chicken carcass and food contact surface samples with retail store risk categorization. The Chi-square test was performed to evaluate statistical differences in the percentage of *C. jejuni*-positive and *C. coli*-positive carcass and food contact surface samples. All statistical analyses were performed with XLSTAT 2018.4, with a significance of $p < 0.05$.

The prevalence of *Campylobacter*-positive carcass samples was 94.0 (47/50) and 32.0% (16/50) in kosher and conventional abattoirs, respectively ($p < 0.0001$). The discrimination of results according to *Campylobacter* species showed 13/50 (26.0%) *C. jejuni*-positive and two *C. coli*-positive carcasses, while only one was positive for both species in the conventional abattoir. In the kosher abattoir, 18 carcasses were positive for *C. jejuni*, one for *C. coli* and 28 for both. The proportion of samples contaminated simultaneously with *C. jejuni* and *C. coli* during kosher and conventional slaughter was 56.0 (28/50) and 2.0% (1/50), respectively ($p < 0.0001$).

Risk categorization of retail stores resulted in 11/25 (44.0%) high-risk, 11/25 (44.0%) moderate-risk and 3/25 (12.0%) low-risk stores. The prevalence of *Campylobacter* spp. in each type of sample from the retail stores is shown in Table 1. From the total number of samples ($n = 25$ each), 19 (76.0%) chicken carcass, 20 (80.0%) table surface and 18

Table 1 Prevalence of *Campylobacter* spp., *Campylobacter jejuni*, *C. coli* and *C. jejuni* + *C. coli* in retail stores.

| | <i>Campylobacter</i> ^a (%) | <i>C. jejuni</i> ^a (%) | <i>C. coli</i> ^a (%) | <i>C. jejuni</i> + <i>C. coli</i> ^a (%) |
|----------------------|--|--------------------------------------|------------------------------------|---|
| | <i>Type of sample</i> | | | |
| Carcass ^b | 19 (76.0) | 4 (16.0) | 8 (32.0) | 7 (28.0) |
| Table ^b | 20 (80.0) | 11 (44.0) | 6 (24.0) | 3 (12.0) |
| Knife ^b | 18 (72.0) | 9 (36.0) | 5 (20.0) | 4 (16.0) |

^a Number of samples with positive isolation. The percentage was calculated over the total number of samples obtained from each type.

^b No statistical differences were observed in the percentage of *C. jejuni*- and *C. coli*-positive samples.

(72.0%) knife surface samples were *Campylobacter*-positive, distributed as follows: one (9.1%) food contact surface and 10 (90.9%) food contact surface and chicken carcass samples in high-risk retail stores; four (33.4%) food contact surface and eight (66.6%) food contact surface and chicken carcass samples in moderate-risk retail stores; one (50.0%) food contact surface and one (50.0%) chicken carcass sample in low-risk retail stores. We found no association between the proportion of samples positive for *Campylobacter* spp. in chicken carcasses ($p=0.180$), food contact surfaces (at least one positive; $p=0.487$) and tables ($p=0.661$) with the risk assigned to retail stores. However, such proportion was higher in knife samples from high-risk retail stores ($p=0.020$).

Undercooked meat and meat products, mainly chicken meat, are most frequently associated with human campylobacteriosis¹³. All stages – from primary broiler production to the consumer – play an important role in the transmission of this pathogen¹³. In our study, the prevalence of *Campylobacter* spp. in chicken carcasses obtained by conventional slaughter (32.0%) was similar to that reported in China (34.1%)⁹ and Argentina (33.3%)¹⁵, but lower than that found in France (96.9%)¹. In agreement with previous studies in Argentinian conventional abattoirs, *C. jejuni* was the most prevalent species isolated from the carcasses¹⁵. While there are no studies on the presence of *Campylobacter* spp. in kosher abattoirs, we observed significant differences in the proportion of *Campylobacter*-positive samples obtained by conventional (32.0%) and kosher (94.0%) slaughter processes, and of samples contaminated simultaneously with *C. coli* and *C. jejuni*. It is important to note some differences between kosher and conventional slaughter. Kosher slaughter must be performed individually by a trained expert called shochet. Shochets use a sharp blade called *chalef* to sever the trachea, esophagus, carotid arteries, jugular veins and vagus nerve. After slaughter, the chicken is placed on a conveyor system and left to hang so that the blood drains from the body. Chicken defeathering must be done using cold water. According to Baker et al.², the absence of heat during scalding undoubtedly contributes to the higher *C. jejuni* count in kosher slaughter. The chicken is then soaked, salted and rinsed to take away any remaining visible blood³. By contrast, conventional abattoirs apply procedures and measures aimed at reducing the risk of microbial contamination of the product, such as desensitization and automated slaughter, defeathering

with hot water (explicitly prohibited in kosher slaughter), automatic gutting, self-cleaning and self-sanitizing, and strict temperature and chlorine control of the chiller equipment¹¹.

Giombelli and Gloria⁷ have shown 100% *Campylobacter* spp. prevalence in broilers from Brazil. The significantly different prevalence between conventional and kosher abattoirs reported in the present study is associated with the slaughter process. The particular characteristics of each method, the slaughter technique used and the risk-mitigation strategies applied to avoid the contamination of meat and the environment with pathogenic bacteria will determine food safety. Therefore, if contamination decreases in the abattoirs, it would be possible to prevent it from reaching the retail stores.

In this study, the prevalence of *Campylobacter* spp. in retail store chicken carcasses (76.0%) agrees with previously reported data in Argentina (83.0%)¹⁵. However, it was higher than that found in China (31.3%)⁹ and in another study conducted in Argentina⁶. While *C. coli* was most frequently isolated from broiler carcasses, *C. jejuni* was isolated from food contact surfaces. The higher presence of *C. coli* regarding *C. jejuni* in chicken carcasses from the retail stores detected in this work differs from data of previous studies from Argentina^{6,15}, USA¹⁴ and Europe¹⁰. However, *C. coli* was the most prevalent species isolated from chicken liver in Argentina¹⁵ and Chile⁵. Although laboratory methods could facilitate the isolation of one species over the other, differences in prevalence could be due to the risk categorization of retail stores. The retail environment can be the source of food contamination with pathogenic bacteria, and the absence of standardized sanitation operating procedures would worsen and perpetuate this situation^{8,12}.

Based on our results, retail stores and abattoirs were sources of chicken carcass contamination with *Campylobacter* spp. Differences in the proportion of chicken carcasses contaminated with *Campylobacter* spp. and simultaneously with *C. jejuni* and *C. coli* in kosher vs. conventional abattoirs were significant. *C. coli* was the species most frequently isolated from chicken carcasses in retail stores from Argentina. More studies are needed to determine the risk of acquiring a foodborne disease due to kosher chicken consumption. The application of corrective actions on variables associated with the presence of *Campylobacter* spp. in abattoirs, particularly kosher ones, and good manufacturing practices in

retail stores could improve the prevention of thermotolerant *Campylobacter*-caused disease.

Funding sources

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Conflict of interest

None declared.

Acknowledgments

The authors thank A. Di Maggio for manuscript correction and editing.

References

1. Alpigliani I, Abrahantes JC, Michel V, Huneau-Salaün A, Chemaly M, Keeling LJ, Gervelmeyer A, Bacci C, Brindani F, Bonardi S, Berthe F. Associations between animal welfare indicators and *Campylobacter* spp. in broiler chickens under commercial settings: a case study. *Prev Vet Med.* 2017;147:186–93.
2. Baker RC, Paredes MD, Qureshi RA. Prevalence of *Campylobacter jejuni* in eggs and poultry meat in New York State. *Poult Sci.* 1987;66:1766–70.
3. Cor.ca. Toronto: COR. Available from: http://www.cor.ca/view/681/what_is_shechita_kosher_slaughter.html [Internet; cited 15.03.19].
4. EFSA (European Food Safety Authority) and ECDC (European Centre for Disease Prevention and Control). The European Union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2015. *EFSA J.* 2016;14:4634–865.
5. Fernández H, Pisón V. Isolation of thermotolerant species of *Campylobacter* from commercial chicken livers. *Int J Food Microbiol.* 1996;29:75–80.
6. Giacoboni G, Puchuri MC, Cerdá R. *Campylobacter* termotolerantes en menudos y carcasas de pollos provenientes de diferentes comercios de la ciudad de La Plata. *Analecta Vet.* 1999;19:51–4.
7. Giombelli A, Gloria MB. Prevalence of *Salmonella* and *Campylobacter* on broiler chickens from farm to slaughter and efficiency of methods to remove visible fecal contamination. *J Food Prot.* 2014;77:1851–959.
8. Leotta GA, Brusa V, Galli L, Adriani C, Linares L, Etcheverría A, Sanz M, Sucari A, Peral García P, Signorini M. Comprehensive evaluation and implementation of improvement actions in butcher shops. *PLoS ONE.* 2016;11:e0162635.
9. Ma L, Wang Y, Shen J, Zhang Q, Wu C. Tracking *Campylobacter* contamination along a broiler chicken production chain from the farm level to retail in China. *Int J Food Microbiol.* 2014;181:77–84.
10. Osimani A, Aquilanti L, Pasquini M, Clementi F. Prevalence and risk factors for thermotolerant species of *Campylobacter* in poultry meat at retail in Europe. *Poult Sci.* 2017;96:3382–91.
11. Servicio Nacional de Sanidad y Calidad Agroalimentaria. Decreto 4238/68. Capítulo XX. Matadero de Aves.
12. Silva J, Leite D, Fernandes M, Mena C, Gibbs PA, Teixeira P. *Campylobacter* spp. as a foodborne pathogen: a review. *Front Microbiol.* 2011;27:200.
13. Skarp CPA, Hänninen ML, Rautelin HIK. *Campylobacteriosis*: the role of poultry meat. *Clin Microbiol Infect.* 2016;22:103–9.
14. Williams A, Oyarzabal OA. Prevalence of *Campylobacter* spp. in skinless, boneless retail broiler meat from 2005 through 2011 in Alabama, USA. *BMC Microbiol.* 2012;12:184.
15. Zbrun MV, Romero-Scharpen A, Olivero C, Rossler E, Soto LP, Rosmini MR, Sequeira GJ, Signorini ML, Frizzo LS. Occurrence of thermotolerant *Campylobacter* spp. at different stages of the poultry meat supply chain in Argentina. *N Z Vet J.* 2013;61:337–43.