Food Control 50 (2015) 328–330

Contents lists available at ScienceDirect

Food Control

journal homepage: www.elsevier.com/locate/foodcont

Short Communication

Prevalence and antimicrobial resistance of *Campylobacter* species isolates in ducks and geese

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ARTICLE INFO

Article history:
Received 24 March 2014
Received in revised form
5 September 2014
Accepted 13 September 2014
Available online 22 September 2014

Keywords:
Campylobacteriosis
Campylobacter
Duck
Goose
Antimicrobial resistance

ABSTRACT

A total of 471 duck and goose intestinal content samples were collected from wet markets and were determined for the prevalence of *Campylobacter* spp. For the detected isolates, resistance to selected antimicrobial agents was identified. *Campylobacter* spp. was detected in 114/291 duck samples (39.2%) and 47/180 goose samples (26.1%). Among the 161 isolated *Campylobacter* spp., 85.7% and 14.3% were *Campylobacter jejuni* and *Campylobacter coli*, respectively. Resistance to ciprofloxacin (82.6%), tetracycline (77%) and nalidixic acid (75.2%) was particularly high in the tested *Campylobacter* isolates. However, all isolates were susceptible to gentamicin, neomycin, streptomycin, neomycin and ampicillin. The presence of *Campylobacter* spp., as well as the detection of multidrug-resistant isolates in this study, indicates that consuming of duck and goose meat might be a potential campylobacteriosis risk in this region.

1. Introduction

Campylobacter is an important pathogen that causes sporadic human foodborne illness and economic losses globally. Isolating of *Campylobacter* from the gastrointestinal tract of poultry and pigs in previous studies showed that these animals can be reservoirs for the pathogen (Allen et al., 2007; Oosterom, De Wilde, De Boer, De Blaauw, & Karman, 1983). The two significant types of *Campylobacter* species associated with serious health problems in humans are *Campylobacter jejuni* and *Campylobacter coli* (Wesley et al., 2000).

The occurrence of antimicrobial-resistant foodborne pathogens is an emerging issue in both human and veterinary medicine. This is due to the over-dependency of antimicrobial agents that cause the emergence, selection and spread of antimicrobial resistance among pathogenic bacteria (Sayah, Kaneene, Johnson, & Miller, 2005). The development of antimicrobial resistance in *Campylobacter* spp. is a serious issue, described in several studies as a problem to public health. The problem arises from the indiscriminate application of antimicrobials in animal products, which leads to the spread of antimicrobial-resistant *Campylobacter* isolates (Adzitey, Rusul, Huda, Cogan, & Corry, 2012; Silva et al., 2011). One of the major sources of *Campylobacter* infection is contaminated poultry meat, and therefore applying strategies to reduce *Campylobacter* in the host would assist with the reduction of *Campylobacter*-associated incidences (Janssen et al., 2008; Riazi et al., 2013).

The objectives of the current study were to investigate the prevalence of *Campylobacter* spp. and their susceptibility to selected antimicrobials in duck and goose intestinal content samples in Varamin, Tehran Province, Iran.

2. Materials and methods

2.1. Sample collection

Between November 2008 and July 2010, 291 duck and 180 goose intestinal content samples were sampled from wet markets in Varamin, Tehran Province, Iran. The samples were transferred into sterile plastic bags and transported in icebox to the laboratory within 3 h of sampling.

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http://dx.doi.org/10.1016/j.foodcont.2014.09.016
0956-7135/

Table 1
Prevalence (% in parentheses) of *Campylobacter* spp. in duck and goose samples.

Source	Total	*Campylobacter* spp.	*Campylobacter jejuni*	*Campylobacter coli*
Duck	291	114 (39.2)	102 (89.5)	12 (10.5)
Goose	180	47 (26.1)	36 (76.6)	11 (23.4)
Total	471	161 (34.2)	138 (29.3)	23 (4.9)

2.2. Isolation and identification of Campylobacter spp.

Campylobacter spp. isolation was carried out following by ISO 10272-1 (ISO, 2006). Briefly, 25 g of each intestinal content sample was weighted and mixed with 225 ml of Preston broth (Oxoid, Basingstoke, UK) and incubated for 24–48 h at 42 °C under anaerobic condition using commercial gas packs (Oxoid Campygen) in microaerophilic atmosphere. The enrichment broth was then streaked on Preston agar as selective medium and incubated at 42 °C in jar for 48 h. Several presumptive *Campylobacter* colonies were picked from each plate and subjected to biochemical tests. Finally, the API Campy identification system (Biomerieux) was used to identify and differentiate of the *Campylobacter* species.

2.3. Antimicrobial susceptibility test

Antimicrobial susceptibility was determined by the Kirby–Bauer disc diffusion method with Mueller Hinton agar (Oxoid, Basingstoke, UK) supplemented with 5% horse blood (Oxoid, Basingstoke, UK) (CLSI, 2006, 2008). Amoxicillin (30 μg), ampicillin (30 μg), nalidixic acid (30 μg), chloramphenicol (30 μg), ciprofloxacin (5 μg), colistin (10 μg), erythromycin (15 μg), gentamicin (10 μg), neomycin (30 μg), streptomycin (30 μg) and tetracycline (15 μg) were used as antimicrobial agents in this study.

2.4. Statistical analysis

Chi-square analysis was carried out to investigate the relationship between different kinds of normal and contaminated samples. SPSS 18.0 (SPSS Inc., Chicago, IL, USA) was applied for all statistical and chi-square analyses. A P value of <0.05 was employed for statistical significance.

3. Results

The *Campylobacter* spp. prevalence rates in duck and goose intestinal content samples are summarized in Table 1. Overall, 161 samples (34.2%) were contaminated with *Campylobacter* spp. in this study. The pathogen was isolated in 39.2% and 26.1% of duck and goose samples, respectively. *C. jejuni* (85.7%) was the most prevalent *Campylobacter* spp. isolated from the samples and the remaining serovar was *C. coli* (14.3%).

The antimicrobial resistance profiles for 138 *C. jejuni* and 23 *C. coli* isolates are presented in Table 2. Out of 161 *Campylobacter* isolates, 156 (96.9%) demonstrated resistance to one or more antimicrobial agents. Twenty (12.4%) and 39 (24.2%) isolates showed resistance to only one and two antimicrobial agents, respectively. Moreover, 97 isolates (60.2%) were multidrug-resistant (to three or more antimicrobial agents). The isolates were resistant to ciprofloxacin (86.8%), tetracycline (75.4%) and nalidixic acid (74.6%). All *Campylobacter* isolates were sensitive to gentamicin. However, all *C. coli* isolates were also susceptible to chloramphenicol. *C. coli* isolates were generally more susceptible to antimicrobial agents than *C. jejuni* isolates.

4. Discussion

According to the findings, there was high prevalence of *Campylobacter* spp. in duck (39.2%) and goose (26.1%) intestinal content; however, there was no significant difference between duck and goose samples. Poultry intestinal tract is considered a favorable environment for *Campylobacter* colonization (Mirzaie, Hassanzadeh, Bashashati, & Barrin, 2011). Poultry meat contamination by foodborne pathogens may occur during carcass processing (Rahimi & Ameri, 2011) and pose public health risks.

The results indicate that *Campylobacter* spp. is highly prevalence in duck and goose, which is similar to earlier findings regarding duck and goose samples in Iran (Rahimi, Alian, & Alian, 2011). However, the prevalence of *Campylobacter* spp. in duck samples in the current study was less than previous reports from the UK (Colles, Ali, Sheppard, McCarthy, & Maiden, 2011), Malaysia (Adzitey et al., 2012), Taiwan (Tsai & Hsiang, 2005), Tanzania (Nonga & Muhairwa, 2010) and Nigeria (Akwuobu & Ofukwu, 2010). Nonetheless, McCrea et al. (2006) reported a higher percentage of contaminated duck samples in the USA. The differences among results might be due to diverse sampling techniques, geographic and

Table 2
Number and percentages of antimicrobial resistance of *Campylobacter* spp. isolated from duck and goose samples.

	Resistant breakpoint (mm)	*Campylobacter* spp.			*Campylobacter jejuni*			*Campylobacter coli*		
		Duck (114)	Goose (47)	Total (161)	Duck (102)	Goose (36)	Total (138)	Duck (12)	Goose (11)	Total (23)
Amoxicillin	≤13	35 (30.7%)	12 (25.5%)	47 (29.2%)	33 (32.4%)	11 (30.6%)	44 (31.9%)	2 (16.7%)	1 (9.1%)	3 (13%)
Ampicillin	≤13	14 (12.3%)	2 (4.2%)	16 (9.9%)	13 (12.7%)	2 (5.6%)	15 (10.9%)	1 (8.3%)	0	1 (4.3%)
Chloramphenicol	≤12	5 (4.4%)	1 (2.1%)	6 (3.7%)	5 (4.9%)	1 (2.8%)	6 (4.3%)	0	0	0
Ciprofloxacin	≤15	99 (86.8%)	34 (72.3%)	133 (82.6%)	91 (89.2%)	27 (75%)	118 (85.5%)	8 (66.7%)	7 (63.6%)	15 (65.2%)
Colistin	≤10	27 (23.7%)	13 (27.7%)	40 (24.8%)	22 (21.6%)	10 (27.8%)	32 (23.2%)	5 (41.7%)	3 (27.3%)	8 (34.8%)
Erythromycin	≤13	5 (4.4%)	1 (2.1%)	6 (3.7%)	4 (3.9%)	1 (2.8%)	5 (3.6%)	1 (8.3%)	0	1 (4.3%)
Gentamicin	≤12	0	0	0	0	0	0	0	0	0
Neomycin	≤12	4 (3.5%)	0	4 (2.5%)	3 (2.9%)	0	3 (2.2%)	1 (8.3%)	0	1 (4.3%)
Streptomycin	≤12	4 (3.5%)	0	4 (2.5%)	3 (2.9%)	0	3 (2.2%)	1 (8.3%)	1	1 (4.3%)
Tetracycline	≤14	86 (75.4%)	38 (80.9%)	124 (77%)	79 (77.5%)	30 (83.3%)	109 (79%)	7 (58.3%)	8 (72.7%)	15 (65.2%)
Nalidixic acid	≤13	85 (74.6%)	36 (76.6%)	121 (75.2%)	74 (72.5%)	29 (80.6%)	103 (74.6%)	11 (91.7%)	7 (63.6%)	18 (78.3%)
R to 1 a.m.[a]	–	16 (14%)	4 (8.5%)	20 (12.4%)	14 (13.7%)	3 (8.3%)	17 (12.3%)	2 (16.7%)	1 (9.1%)	3 (13%)
R to 2 a.m.[b]	–	25 (21.9%)	14 (29.8%)	39 (24.2%)	22 (21.6%)	8 (22.2%)	30 (21.7%)	3 (25%)	6 (54.5%)	9 (39.1%)
R to >2 a.m.[c]	–	70 (61.4%)	27 (57.4%)	97 (60.2%)	64 (62.7%)	23 (63.9%)	87 (63%)	6 (50%)	4 (36.4%)	10 (43.5%)

[a] R to 1 a.m.: resistance to 1 antimicrobial.
[b] R to 2 a.m.: resistance to 2 antimicrobials.
[c] R to >2 a.m.: resistance to >2 antimicrobials.

seasonal effects (Willis & Murray, 1997) as well as laboratory techniques employed in each study (Rahimi & Ameri, 2011).

The most predominate serovar was *C. jejuni*, which has been reported in food of animal origin especially poultry meat (Adzitey et al., 2012; Colles et al., 2011; Nonga & Muhairwa, 2010; Tsai & Hsiang, 2005; Whyte et al., 2004), whereas *C. coli* was predominant in pigs (Rosef, Gondrosen, Kapperud, & Underdal, 1983; Thakur & Gebreyes, 2005).

Our results are in agreement with earlier studies (Little, Richardson, Owen, De Pinna, & Threlfall, 2008; Rahimi & Ameri, 2011; Soltan Dallal et al., 2010; Zendehbad, Arian, & Alipour, 2013), which reported a high prevalence of *Campylobacter* resistant to quinolones (nalidixic acid), fluoroquinolones (ciprofloxacin), and tetracyclines (tetracycline) among duck and goose samples. The high prevalence of ciprofloxacin- and tetracycline-resistant isolates in the current study and past findings in Iran (Rahimi & Ameri, 2011; Soltan Dallal et al., 2010; Zendehbad et al., 2013) could be attributable to the widespread use of these antimicrobial agents in the last decades for the treatment, control and prevention of poultry diseases (Mirzaie et al., 2011; Rahimi & Ameri, 2011). Therefore, using antimicrobial agents in poultry rearing may lead to the emergence of antimicrobial-resistant *Campylobacter* in treated birds (Luangtongkum et al., 2009).

All *Campylobacter* spp. were susceptible to gentamicin and the outcome was similar to other reported studies (Chen et al., 2010; Rahimi et al., 2011; Rahimi & Ameri, 2011; Soltan Dallal et al., 2010; Zendehbad et al., 2013). The low levels of gentamicin-resistant isolates detected are potentially owing to less use of this antimicrobial agent in duck and goose rearing. In the present study, 29.2% and 3.7% of *Campylobacter* isolates were resistant to amoxicillin (beta-lactam) and erythromycin (macrolide), respectively. However, all tested *Campylobacter* isolates were susceptible to amoxicillin and erythromycin in earlier findings on duck and goose samples in Iran (Rahimi et al., 2011). Furthermore, our results illustrate high levels of multidrug resistance among the isolated *Campylobacter* spp. The presence of multidrug-resistant isolates could be a significant threat to human health as the types of effective antibiotics become limited (Chen et al., 2010; Han, Jang, Choo, Heu, & Ryu, 2007; Rahimi & Ameri, 2011; Zendehbad et al., 2013).

In summary, the presence of *Campylobacter* spp. and detection of multidrug-resistant isolates in duck and goose intestinal content in this study indicate that consuming these types of meat is a potential public health risk regarding foodborne campylobacteriosis. The surveillance of *Campylobacter* spp. and stringent monitoring of antimicrobial agent usage in poultry rearing would reduce the risk of meat contamination and development of antibiotic-resistant isolates.

Acknowledgments

This study was supported by OCAR chancellery of University Malaya with grant number A-21010-DA674 and A-21010-DA677.

References

Adzitey, F., Rusul, G., Huda, N., Cogan, T., & Corry, J. (2012). Prevalence, antibiotic resistance and RAPD typing of *Campylobacter* species isolated from ducks, their rearing and processing environments in Penang, Malaysia. *International Journal of Food Microbiology, 154*(3), 197–205.

Akwuobu, C., & Ofukwu, R. (2010). *Campylobacter jejuni* in duck faeces around drinking water sources in Makurdi, North-Central Nigeria: public health implication. *Tanzania Veterinary Journal, 27*(1), 21–26.

Allen, V., Bull, S., Corry, J., Domingue, G., Jørgensen, F., Frost, J., et al. (2007). *Campylobacter* spp. contamination of chicken carcasses during processing in relation to flock colonisation. *International Journal of Food Microbiology, 113*(1), 54–61.

Chen, X., Naren, G.-W., Wu, C.-M., Wang, Y., Dai, L., Xia, L.-N., et al. (2010). Prevalence and antimicrobial resistance of *Campylobacter* isolates in broilers from China. *Veterinary Microbiology, 144*(1), 133–139.

Clinical and Laboratory Standards Institute (CLSI). (2006). *Methods for antimicrobial dilutions and disk susceptibility testing of infrequently isolated or fastidious bacteria: approved Guideline M45-A*. Wayne, PA: Clinical and Laboratory Standards Institute.

Clinical and Laboratory Standards Institute (CLSI). (2008). *Performance standards for antimicrobial disk and dilution susceptibility tests for bacteria isolated from animals: approved guideline M31–A3*. Wayne, PA: Clinical and Laboratory Standards Institute.

Colles, F. M., Ali, J. S., Sheppard, S. K., McCarthy, N. D., & Maiden, M. C. (2011). *Campylobacter* populations in wild and domesticated Mallard ducks (*Anas platyrhynchos*). *Environmental Microbiology Reports, 3*(5), 574–580.

Han, K., Jang, S. S., Choo, E., Heu, S., & Ryu, S. (2007). Prevalence, genetic diversity, and antibiotic resistance patterns of *Campylobacter jejuni* from retail raw chickens in Korea. *International Journal of Food Microbiology, 114*(1), 50–59.

ISO. (2006). *Microbiology of food and animal feed stuffs-horizontal method for detection and enumeration of Campylobacter spp*. Geneva: ISO.

Janssen, R., Krogfelt, K. A., Cawthraw, S. A., van Pelt, W., Wagenaar, J. A., & Owen, R. J. (2008). Host-pathogen interactions in *Campylobacter* infections: the host perspective. *Clinical Microbiology Reviews, 21*(3), 505–518.

Little, C. L., Richardson, J. F., Owen, R. J., De Pinna, E., & Threlfall, E. J. (2008). *Campylobacter* and *Salmonella* in raw red meats in the United Kingdom: prevalence, characterization and antimicrobial resistance pattern, 2003–2005. *Food Microbiology, 25*(3), 538–543.

Luangtongkum, T., Jeon, B., Han, J., Plummer, P., Logue, C. M., & Zhang, Q. (2009). Antibiotic resistance in *Campylobacter*: emergence, transmission and persistence. *Future Microbiology, 4*, 189–200.

McCrea, B., Tonooka, K., VanWorth, C., Boggs, C., Atwill, E., & Schrader, J. (2006). Prevalence of *Campylobacter* and *Salmonella* species on farm, after transport, and at processing in specialty market poultry. *Poultry Science, 85*(1), 136–143.

Mirzaie, S., Hassanzadeh, M., Bashashati, M., & Barrin, A. (2011). *Campylobacter* occurrence and antimicrobial resistance in samples from ceca of commercial turkeys and quails in Tehran, Iran. *International Research Journal of Microbiology, 2*, 338–342.

Nonga, H. E., & Muhairwa, A. (2010). Prevalence and antibiotic susceptibility of thermophilic *Campylobacter* isolates from free range domestic duck (Cairina moschata) in Morogoro municipality, Tanzania. *Tropical Animal Health and Production, 42*(2), 165–172.

Oosterom, J., De Wilde, G., De Boer, E., De Blaauw, L., & Karman, H. (1983). Survival of *Campylobacter jejuni* during poultry processing and pig slaughtering. *Journal of Food Protection, 46*, 702–706.

Rahimi, E., Alian, F., & Alian, F. (2011). Prevalence and characteristic of *Campylobacter* species isolated from raw duck and goose meat in Iran. In *International Conference on Food Engineering and Biotechnology* (vol. 9, pp. 171–175). Singapore: IACSIT Press.

Rahimi, E., & Ameri, M. (2011). Antimicrobial resistance patterns of *Campylobacter* spp. isolated from raw chicken, turkey, quail, partridge, and ostrich meat in Iran. *Food Control, 22*(8), 1165–1170.

Riazi, A., Strong, P. C., Coleman, R., Chen, W., Hirama, T., van Faassen, H., et al. (2013). Pentavalent single-domain antibodies reduce *Campylobacter jejuni* motility and colonization in chickens. *PLoS One, 8*(12), e83928.

Rosef, O., Gondrosen, B., Kapperud, G., & Underdal, B. (1983). Isolation and characterization of *Campylobacter jejuni* and *Campylobacter coli* from domestic and wild mammals in Norway. *Applied and Environmental Microbiology, 46*(4), 855–859.

Sayah, R. S., Kaneene, J. B., Johnson, Y., & Miller, R. (2005). Patterns of antimicrobial resistance observed in Escherichia coli isolates obtained from domestic- and wild-animal fecal samples, human septage, and surface water. *Applied and Environmental Microbiology, 71*(3), 1394–1404.

Silva, J., Leite, D., Fernandes, M., Mena, C., Gibbs, P. A., & Teixeira, P. (2011). *Campylobacter* spp. as a foodborne pathogen: a review. *Frontiers in Microbiology, 2*.

Soltan Dallal, M. M., Doyle, M. P., Rezadehbashi, M., Dabiri, H., Sanaei, M., Modarresi, S., et al. (2010). Prevalence and antimicrobial resistance profiles of Salmonella serotypes, *Campylobacter* and *Yersinia* spp. isolated from retail chicken and beef, Tehran, Iran. *Food Control, 21*(4), 388–392.

Thakur, S., & Gebreyes, W. A. (2005). Prevalence and antimicrobial resistance of *Campylobacter* in antimicrobial-free and conventional pig production systems. *Journal of Food Protection, 68*(11), 2402–2410.

Tsai, H.-J., & Hsiang, P.-H. (2005). The prevalence and antimicrobial susceptibilities of *Salmonella* and *Campylobacter* in ducks in Taiwan. *The Journal of Veterinary Medical Science, 67*(1), 7–12.

Wesley, I., Wells, S., Harmon, K., Green, A., Schroeder-Tucker, L., Glover, M., et al. (2000). Fecal shedding of Campylobacter and *Arcobacter* spp. in dairy cattle. *Applied and Environmental Microbiology, 66*(5), 1994–2000.

Whyte, P., McGill, K., Cowley, D., Madden, R., Moran, L., Scates, P., et al. (2004). Occurrence of *Campylobacter* in retail foods in Ireland. *International Journal of Food Microbiology, 95*(2), 111–118.

Willis, W. L., & Murray, C. (1997). *Campylobacter jejuni* seasonal recovery observations of retail market broilers. *Poultry Science, 76*(2), 314–317.

Zendehbad, B., Arian, A. A., & Alipour, A. (2013). Identification and antimicrobial resistance of *Campylobacter* species isolated from poultry meat in Khorasan province, Iran. *Food Control, 32*(2), 724–727.