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Prevalence and characteristics of *Salmonella* and *Campylobacter* in retail poultry meat in Japan

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SUMMARY

This study was undertaken to determine the prevalence, antimicrobial susceptibility, and genetic relatedness of *Salmonella enterica* subsp. *enterica* and *Campylobacter* spp. in poultry meat, and to analyze the association of genetic types of these bacteria with their geographical distribution and antimicrobial resistance profiles. Fifty-four and 71 of 100 samples were found to be contaminated with *Salmonella* and *Campylobacter*, respectively. Nine *Salmonella* serotypes were found, including *S. enterica* subsp. *enterica* serovar Infantis (33%), Schwarzengrund (12%), Manhattan (9%), and others. *Campylobacter jejuni* and *C. coli* were detected from 64 (64%) and 14 (14%) samples, respectively. *S. enterica* subsp. *enterica* exhibited a high frequency of resistance against tetracycline (78.3%) and streptomycin (68.3%). *C. jejuni* and *C. coli* isolates were resistant to sulfamethoxazole/trimethoprim (90.5%), nalidixic acid (47.3%), ampicillin (45.9%), and ciprofloxacin (40.5%). Cluster analysis was performed for the *Salmonella* isolates using PFGE and for the *Campylobacter* isolates with PFGE as well as comparative genomic fingerprinting, which were combined with the drug-resistance data to analyze with respect to the locations in which the poultry meat was produced. This analysis revealed that *C. jejuni* strains with a particular genotype and antimicrobial resistance profile are spreading in specific areas in Japan.
INTRODUCTION

*Salmonella* and *Campylobacter* are recognized as the major causative agents of human gastroenteritis worldwide (1). In Japan, *Campylobacter* and *Salmonella* are the second and the third most common causes of foodborne illness, respectively, after norovirus. The major route of *Salmonella* and *Campylobacter* infection in humans is through ingestion of contaminated raw or undercooked meat, especially poultry meat (2,3).

Several molecular typing methods have been employed to analyze the epidemiological features of infections caused by *Salmonella* or *Campylobacter*. Pulsed-field gel electrophoresis (PFGE) is the most common method used for molecular typing of *Salmonella* and *Campylobacter*. In the present study, we used PFGE to analyze both *Salmonella* and *Campylobacter* isolates, and also used the comparative genomic fingerprinting (CGF) method of 40-gene CGF (CGF40) for analysis of the *Campylobacter* isolates. The CGF method was developed by Canadian researchers as a high-resolution comparative genomics-based method specific for *C. jejuni* subtyping with high discriminatory power, that is also rapid, low cost, and easily deployable for routine epidemiologic surveillance and outbreak investigations (4).
Previous studies have reported the prevalence and antimicrobial susceptibilities of *Salmonella* and *Campylobacter* in Japan (5,6). However, there is a paucity of data concerning the association of genetic types of *Salmonella* and *Campylobacter* with their geographical distribution and antimicrobial resistance profiles. Furthermore, the CGF method has not been applied to analyze *Campylobacter* strains isolated in Japan. The aim of this study was to determine the prevalence and antimicrobial susceptibility of *Salmonella* and *Campylobacter* in retail broiler meat, and to describe the molecular characteristics of these pathogens isolated from poultry meat sold in Japan, by using PFGE for *Salmonella* and *Campylobacter* and CGF40 for *Campylobacter*. The results of cluster analysis were combined with data on antimicrobial resistance profiles and further compared with respect to the locations in which the poultry meat was produced.

**MATERIALS AND METHODS**

**Collection of chicken meat samples:** Twenty samples of poultry meat were purchased directly by each of five institutes participating in this study from retail stores from July to October 2012 in the local areas of each institute. Meats of 32 packages were produced in the eastern area (1 package in the Hokkaido region and 31 packages in the Tohoku region), and 49 were in the western area (17 in the Kinki region, 3 in the
Shikoku region and 29 in the Kyushu region) (Table 1). The production area of 19 meat packages was indicated as “domestic”. The meat samples were kept in a refrigerator at 4°C until use, and all samples were processed within one day of purchase.

**Isolation of Salmonella:** Isolation of *Salmonella* was performed by each institute, according to the National Standard Method for *Salmonella* (7). In brief, 25 g of the meat sample was mixed with 225 ml of buffered peptone water. After incubation at 36–38°C for 20–22 h, 1 ml and 0.1 ml of buffered peptone water culture was transferred into 10 ml of tetrathionate broth and 10 ml of Rappaport-Vassiliadis broth, respectively, and incubated at 41.5–42.5°C for 20–24 h. The culture from each tube was inoculated onto DHL agar plates (Nissui Pharma, Tokyo, Japan) and ES *Salmonella* agar II plates (Eiken Chemical, Tokyo, Japan), and incubated at 36–38°C for 20–24 h. Three suspected colonies were chosen from each plate and subjected to biochemical examination for identifying *Salmonella*. Serotyping of *Salmonella* isolates was performed using commercial O and H antisera (Denka Seiken, Tokyo, Japan) and the isolates were designated according to the Kauffmann-White scheme (8).

**Isolation of Campylobacter:** Isolation of *Campylobacter* was performed by each
institute, according to the National Standard Method for *C. jejuni* and *C. coli* (9). In brief, 25 g of a meat sample was mixed with 100 ml of Preston broth and incubated under microaerobic conditions (7–13% CO₂ and 3–7% O₂) at 41–43°C for 24–48 h. After incubation, the culture was inoculated onto modified charcoal cefoperazone deoxycholate agar plates (Eiken Chemical) and Butzler’s selective agar plates (Oxoid, Basingstoke, UK), and the plates were incubated under microaerobic conditions at 41–43°C for 24–48 h. Suspected colonies were presumptively identified as *Campylobacter* on the basis of morphology, Gram-negative staining, and a positive catalase and oxidase reaction. Indoxyl acetate and hippurate hydrolysis tests were conducted to confirm the *Campylobacter* species level. In addition, a PCR assay was utilized to identify *C. jejuni* and *C. coli* (10).

**Antimicrobial susceptibility test:** The antimicrobial susceptibility test was performed based on the Kirby-Bauer disc diffusion method using BBL Sensi-disc susceptibility test discs (BD, Tokyo, Japan) on Mueller-Hinton II agar (BD) for *Salmonella* and on Mueller-Hinton II agar with 5% lysed horse blood for *Campylobacter*. The results were interpreted according to Clinical and Laboratory Standards Institute criteria (11). The results of *Campylobacter* were interpreted according to the criteria for
Enterobacteriaceae. Escherichia coli ATCC25922 was used as a quality control strain.

**PFGE:** PFGE profiles of Salmonella and Campylobacter isolates were determined using a previously described protocol (12) with minor modifications. Bacterial cells on an agar medium were directly embedded in agarose plugs. DNA was digested with 40 U of XbaI or BlnI (Takara Bio Inc., Shiga, Japan) for 5 h at 37°C for Salmonella isolates, and with 40 U of Smal for 5 h at 30°C for Campylobacter isolates. Salmonella enterica subsp. enterica serovar Braenderup H9812 was used as the control strain. PFGE was performed on a 1% agarose gel using a CHEF MAPPER apparatus (Bio-Rad, Tokyo, Japan). The stained gels with ethidium bromide (0.5μg/ml) were observed under UV transillumination.

**CGF method:** The CGF40 method was performed on C. jejuni isolates and C. coli isolates, according to Taboada et al. (4). The PCR mixture consisted of 1U Takara Ex Taq DNA polymerase (Takara Bio), 1X PCR buffer (Takara Bio), 2.5 mM MgCl2, 0.2 mM each deoxynucleoside triphosphate (dNTP), 0.4 M each of the 10 primers, and 1 μl DNA template in a 25-μl reaction mixture. The PCR condition consisted of an initial denaturation of 5 min at 95°C, 35 cycles of 30 s at 94°C, 60 s at 55°C, 60 s at 72°C, a
final extension step of 5 min at 72°C, and a hold at 4°C. The PCR product was visualized by ethidium bromide staining after electrophoresis on a 2.0% agarose gel. *C. jejuni* strains NCTC 11168 and RM1221 were used as control strains.

**Simpson’s diversity index:** Simpson’s diversity index was used for assessing the discriminatory power of PFGE and CGF40 for *C. jejuni*, and evaluating usefulness of CGF40. The index was calculated through the following formula:

\[
D = 1 - \frac{1}{N(N-1)} \sum_{j=1}^{s} n_j(n_j-1)
\]

where \(N\) was the total number of strains, \(s\) was the total number of patterns, and \(n_j\) was the number of strains that belonged to the \(j\)th pattern (13).

**Cluster analysis:** The PFGE and CGF40 patterns were analyzed using Bionumerics software version 6.6.3 (Applied Maths Sint-Martens-Latem, Belgium). Dendrograms for PFGE and CGF40 patterns were generated based on the unweighted-pair group method using average linkages similarity cluster analysis performed with Dice coefficients, with a 1.0% band position tolerance for PFGE.
**Statistical analysis:** McNemar’s test and Fisher’s exact test were performed using SPSS for Windows release 17.0 software (SPSS Inc., Chicago, USA) to compare the significance of the difference between two groups. Differences of antimicrobial resistance rates and rate of production area between clusters or subclusters with more than five isolates were analyzed with Fisher’s exact test.

**RESULTS**

**Prevalence of *Salmonella* and *Campylobacter* in poultry meat samples:** A total of 100 retail chicken meat samples were purchased in 2012 to determine the prevalence and antibiotic resistance of *Salmonella* and *Campylobacter* spp. The overall prevalence of *Salmonella enterica* subsp. *enterica* and *Campylobacter* spp. from poultry meat is shown in Table 1. The prevalence of *S. enterica* subsp. *enterica* in the meat samples was 54%, consisting of nine serotypes. *S. enterica* subsp. *enterica* serovar Infantis was detected with the highest prevalence, although the prevalence of the different serotypes varied among the areas in which the farms were located.

*Campylobacter* spp. was detected from 71 samples (71%), and *C. jejuni* and *C. coli*
were isolated from 64 (64%) and 14 (14%) meat samples, respectively; seven samples were contaminated with both *C. jejuni* and *C. coli*. *C. lari* was detected from a sample of the thigh.

McNemar’s test was used to test for significant differences between the prevalence of *Salmonella* and *Campylobacter* in paired samples. The results of the test showed a statistically significant difference (*p* < 0.05).

**Antimicrobial susceptibility profiles:** Antimicrobial susceptibility was determined for 60 *S. enterica* subsp. *enterica* isolates (33 isolates of serovar Infantis, 12 of serovar Schwarzengrund, nine of serovar Manhattan, and one each of serovars Agona, Cerro, Corvallis, Duesseldorf, Muenchen, and Typhimurium) and 74 *Campylobacter* isolates (65 isolates of *C. jejuni* and nine of *C. coli*) (Tables 2 and 3). Two strains of *C. jejuni* with different PFGE patterns were isolated from one sample.

Fifty-one *Salmonella* isolates (85.0%) exhibited resistance against at least one antimicrobial agent, and 46 isolates (76.7%) were resistant against more than one antimicrobial agent. The rates of multidrug resistance of *S. enterica* subsp. *enterica* serovars Infantis, Schwarzengrund, and Manhattan were 84.8%, 75.0%, and 88.9%, respectively. One *S. enterica* subsp. *enterica* serovars Infantis strain exhibited drug
resistance against seven antimicrobials: ampicillin, cefotaxime, chloramphenicol, kanamycin, streptomycin, sulfamethoxazole/trimethoprim, and tetracycline. The most frequent multidrug resistance pattern among *S. enterica* subsp. *enterica* serovars *Infantis* isolates was resistant to streptomycin, sulfamethoxazole/trimethoprim, and tetracycline (seven isolates), followed by kanamycin, streptomycin, sulfamethoxazole/trimethoprim, and tetracycline (four isolates), and streptomycin, tetracycline and nalidixic acid (four isolates).

Among the *S. enterica* subsp. *enterica* serovar *Infantis* isolates, three of the five ampicillin-resistant isolates were cefotaxime-resistant, and were isolated from meat samples from the western area. No isolates of *S. enterica* subsp. *enterica* serovar *Infantis* from meat samples from the eastern area exhibited resistance against ampicillin or cefotaxime.

Sixty-four isolates of *C. jejuni* and seven *C. coli* isolates were resistant to at least one antimicrobial agent. Fifty-five *C. jejuni* isolates (84.6%) and five *C. coli* isolates (55.6%) were resistant to more than one antimicrobial agent. Each one strain of *C. jejuni* and *C. coli* was resistant against six antimicrobials: ampicillin, kanamycin, streptomycin, sulfamethoxazole/trimethoprim, nalidixic acid and ciprofloxacin, and ampicillin, kanamycin, tetracycline, sulfamethoxazole/trimethoprim, nalidixic acid and
ciprofloxacin, respectively. The most frequent multidrug resistance pattern among *C. jejuni* was resistant to sulfamethoxazole/trimethoprim, nalidixic acid and ciprofloxacin (six isolates), followed by tetracycline, sulfamethoxazole/trimethoprim, nalidixic acid and ciprofloxacin (five isolates).

**Cluster analysis of Salmonella:** Fifty-three of the *Salmonella* isolates, including 33 isolates of *S. enterica* subsp. *enterica* serovar Infantis, 12 isolates of *S. enterica* subsp. *enterica* serovar Schwarzengrund, and nine isolates of *S. enterica* subsp. *enterica* serovar Manhattan, were subjected to cluster analysis.

*S. enterica* subsp. *enterica* serovar Infantis formed three clusters with a similarity cutoff of 60% (Fig. 1a). Nine isolates in cluster A were resistant to kanamycin (45.0%), while no isolate in cluster B was resistant to kanamycin (Fisher’s exact test: *p* < 0.01). In contrast, four isolates (36.4%) in cluster B were resistant to nalidixic acid, while only one isolate (5.0%) in cluster A was resistant (Fisher’s exact test: *p* < 0.05). No other significant difference in microbial resistance rates or rate of production area between clusters was observed.

The combined *XbaI* and *BlnI* dendrogram showed that each of the studied population of *S. enterica* subsp. *enterica* serovars Schwarzengrund and Manhattan
isolates was genetically similar (Fig. 1b and 1c).

**PFGE and CGF40 for *C. jejuni* strains:** Sixty-five isolates of *C. jejuni* were analyzed using the PFGE method to identify 55 patterns (Fig. 2a). The same 65 isolates of *C. jejuni* were analyzed using CGF40 to identify 49 patterns (Fig. 2b). Simpson’s diversity indices for PFGE and CGF40 for *C. jejuni* were 0.997 and 0.990, respectively.

**Cluster analysis of Campylobacter:**

Sixty-three isolates of *C. jejuni* formed six clusters by PFGE with a similarity of 40% (Fig. 2a). Two *C. jejuni* isolates were not assigned to any cluster. Isolates belonging to cluster P5 were divided into four subclusters with 55% similarity. Resistance rate of isolates in cluster P4 (100%) to sulfamethoxazole/trimethoprim was different from that of isolates in cluster P6 (62.5%, Fisher’s exact test: \( p < 0.05 \)). No isolate in subcluster I was resistant to tetracycline, while five isolates in subcluster II (50.0%, Fisher’s exact test: \( p < 0.05 \)) were resistant to tetracycline. Additionally, resistance rate of isolates in subcluster II (80.0% and 80.0%) to nalidixic acid and ciprofloxacin were significantly different from those of isolates in subcluster I (14.3% and 14.3%, Fisher’s exact test: \( p < 0.05 \)). No other significant difference in microbial
resistance rates or rate of production area between clusters or subclusters was observed.

The 63 C. jejuni isolates formed seven clusters by CGF40 with 65% similarity (Fig. 2b). The remaining two isolates were not assigned to any cluster. It was observed that there were statistically significant differences in microbial resistance rates between clusters as follows: kanamycin resistance between cluster C3 (12.5%) and C5 (57.9%) (Fisher’s exact test: \( p < 0.05 \)), and between cluster C4 (8.3%) and C5 (57.9%) (\( p < 0.01 \)), tetracycline resistance between cluster C4 (66.7%) and C6 (23.5%) (\( p < 0.05 \)), nalidixic acid resistance between cluster C3 (12.5%) and C4 (83.3%) (\( p < 0.01 \)), between cluster C3 (12.5%) and C5 (63.2%) (\( p < 0.05 \)), between cluster C4 (83.3%) and C6 (17.6%) (\( p < 0.01 \)), and between cluster C5 (63.2%) and C6 (17.6%) (\( p < 0.01 \)), and ciprofloxacin resistance between cluster C3 (12.5%) and C4 (83.3%) (\( p < 0.01 \)), between cluster C4 (83.3%) and C5 (47.4%) (\( p < 0.05 \)), between cluster C4 (83.3%) and C6 (11.8%) (\( p < 0.01 \)), and between cluster C5 (47.4%) and C6 (11.8%) (\( p < 0.05 \)). Eight and four strains in cluster C4 were isolated from poultry meat produced in the eastern and western area, respectively, while three strains were from the eastern area and nine strains were from the western area in cluster C6. Difference in rates of production area between these clusters was significant (Fisher’s exact test: \( p < 0.05 \)). No other significant difference in microbial resistance rates or rate of production area between clusters was observed.
In analyses using PFGE and CGF40, all *C. coli* isolates fell into one distinct group from *C. jejuni* (data not shown).

**DISCUSSION**

We determined the prevalence of *Salmonella* and *Campylobacter* in poultry meat purchased in Japan, and found that 54% and 71% of the poultry meat samples were contaminated with *Salmonella enterica* subsp. *enterica* and *Campylobacter* spp., respectively. These frequencies are consistent with those reported in previous studies (14,15), but are higher than those in other reports (5,16–19). Several factors might influence the prevalence of *Salmonella* and *Campylobacter* in poultry meat. Differences in the prevalence of *Salmonella* and *Campylobacter* in poultry meat could be attributed to differences in environmental factors such as the geographical location of farms and the season in which the study is carried out, as well as differences in culture and sampling methods (20).

The prevalence of *Salmonella* in poultry meat was lower than that of *Campylobacter*. In addition, the prevalence of *Salmonella* and *Campylobacter* varied by part of chicken meat with a higher *C. jejuni* prevalence than that of *Salmonella* in tenderloin. The results of McNemar’s test showed that contamination of *Salmonella* and *Campylobacter*
in poultry meat occurred independently. Wilson (21) indicated that there was no significant association between *Salmonella* and *Campylobacter* as co-contaminants in retail chicken meat, and that control measures that have reduced *Salmonella* contamination have not been effective against *Campylobacter*. Cason *et al.* (22) also reported that the contamination of *Salmonella* and *Campylobacter* in chicken carcasses occurred as independent events. Additionally, while *C. jejuni* was detected with 75.0% in tenderloin, *C. coli* was not detected from tenderloin, likely because of low detection rate.

The *Salmonella* isolates analyzed in this study exhibited a high prevalence of antimicrobial resistance to kanamycin, streptomycin, tetracycline, and sulfamethoxazole/trimethoprim. Furthermore, the *C. jejuni* and *C. coli* isolates were highly resistant to sulfamethoxazole/trimethoprim, and approximately 30 to 50% of isolates were resistant to ampicillin, kanamycin, tetracycline, nalidixic acid, and ciprofloxacin. The reasons for the difference in the resistance patterns between the two genera are not currently clear, but may be due to difference in the mechanism for acquisition of the factors that confer drug resistance and/or ecological features.

In the present study, *S. enterica* subsp. *enterica* serovar Infantis was detected in retail poultry meat from all areas, and was predominant in the eastern area. These results
were comparable to other studies, despite multi-year gap between the present study and previous studies. Iwabuchi et al. (19) reported that *S. enterica* subsp. *enterica* serovar *Infantis* exhibited the highest prevalence among detected *Salmonella* serovars in poultry meat purchased in the northern (the Hokkaido region) and central part (the Tokai region) of Japan, but was not isolated from poultry meat purchased in the southern part (the Kyushu region) between December 2006 and March 2008. Sasaki et al. (6) divided Japan into 2 areas: the east (the Hokkaido, Tohoku, Kanto, and Tokai regions) and the west (the Kinki, Chugoku, Shikoku, and Kyushu regions), and demonstrated that prevalence of *S. enterica* subsp. *enterica* serovar *Infantis* in broiler flocks in the east area was higher than that in the west area between November 2007 and February 2010.

In the present study, the *S. enterica* subsp. *enterica* serovars Schwarzengrund and Manhattan showed high prevalence in poultry meat from the western area, while these serotypes were detected with low frequency in poultry meat from the eastern area. The results of low variation in their PFGE patterns and the limited geographical distribution of these serovars suggest that strains with clonal relatedness might be brought into the western area of Japan, and might have since been spreading into other areas. The results of the geographical distribution of the two serovars are consistent with the previous studies (6,19). *S. enterica* subsp. *enterica* serovars Schwarzengrund and Manhattan
were first detected from broiler chicken in the western area of Japan in 2005 and 2007, respectively (23,24). *S. enterica* subsp. *enterica* serovar Schwarzengrund was detected in the eastern area in 2009, suggesting that this serovar spread from the western area into the eastern area of Japan (6). Further studies should be conducted to elucidate other possibilities that *S. enterica* subsp. *enterica* serovar Schwarzengrund strains might be introduced from foreign countries into the eastern area.

Isolates of *C. jejuni* were analyzed to determine genetic similarity using CGF40 in this study. Simpson’s diversity index for CGF40 for 65 *C. jejuni* isolates was comparable to that for PFGE for the same *C. jejuni* isolates. These results suggest that CGF40 is a useful tool for analyzing and determining genetic relatedness of *C. jejuni* in combination with PFGE. This new method is a useful tool for molecular typing for determining the genetic relatedness of *C. jejuni*, and is highly concordant with multilocus sequence typing results, but with better discriminatory power (Carrillo et al., 2012, Taboada et al., 2012).

Cluster analysis was conducted to analyze association of antimicrobial resistance and geographical distribution with genetic groups of strains. The results of cluster analysis for *Salmonella* and *Campylobacter* by PFGE and CGF obtained in this study suggest that drug resistance may be related to genotype and has become dispersed in a
particular genetic group of strains, or that a group of *C. jejuni* strains with drug resistance have spread in a particular geographical area. The degree of association between specific genotypes and drug resistance in *Campylobacter* and *Salmonella* remains a topic of debate. Some researchers reported that drug resistance is associated with specific genotypes (25–29), whereas others reported no such association (30, 31). Because the limited number of isolates was analyzed in this study, further studies should be conducted to elucidate the association between genotypes and drug resistance in *Campylobacter*.

In conclusion, we found that poultry meat retailed in Japan is highly contaminated with *Salmonella* and *Campylobacter*, and that the majority of isolates were resistant to multiple antimicrobials. The results of cluster analysis in the present study suggested an association between genetic types of *Campylobacter* and antimicrobial resistance patterns, and that a particular group of closely related strains tends to spread in particular areas. Because the present study was a cross-sectional study and the number of analyzed isolates was somewhat small, the information obtained is limited. Therefore, a further longitudinal study should be conducted to investigate the dynamics of the distribution of genetically related strains within a serotype or species, geographical distribution, and the acquisition of drug resistance of *Campylobacter* in poultry meat.
This information should help to determine the patterns of spread of bacteria, the development of antibiotic resistance, which should help to determine effective strategies to prevent contamination of poultry meat by *Campylobacter*. Furthermore, the present study indicated that the new method of CGF is a practically useful tool for analyzing and determining the genetic types of the *Campylobacter* strains.

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**Conflicts of interest**

The authors have no potential conflicts of interest to declare.

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FIGURE LEGENDS

Fig. 1. Combined dendrogram of PFGE patterns with *XbaI* and *BlnI* for *Salmonella* Infantis (a), Schwarzengrund (b), and Manhattan (c). Cluster analysis was performed with Bionumerics using the Dice correlation coefficient and the UPGMA clustering algorithm. Black squares indicate resistance to antibiotics. For abbreviations of antibiotics, see Table 2, footnote.

Fig. 2. Dendrograms of PFGE (a) and CGF40 (b) patterns for *Campylobacter jejuni* and *C. coli*. Digestion with *SmaI* was used for PFGE analysis. Cluster analysis was performed with Bionumerics using the Dice correlation coefficient and the UPGMA clustering algorithm. Black squares of the CGF patterns indicate a positive PCR result. Black squares of the antibiogram indicate resistance to antibiotics. For abbreviations of antibiotics, see Table 3, footnote. NA: not assigned to any cluster or subcluster.
Table 1. Prevalence of *Salmonella enterica* subsp. *enterica*, *Campylobacter jejuni* and *Campylobacter coli* in retail poultry meat.

<table>
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<th><em>Salmonella enterica</em> subsp. <em>enterica</em></th>
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</tr>
</thead>
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<td>serotypes (^1)</td>
<td></td>
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</tr>
<tr>
<td></td>
<td>chten (54.0)(^2)</td>
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<td>C. jejuni</td>
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<td>C. coli</td>
</tr>
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<td>33 (33.0)</td>
<td>12 (12.0)</td>
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<tr>
<td>Total</td>
<td>100</td>
<td>54 (54.0)(^2)</td>
<td>33 (33.0)</td>
<td>12 (12.0)</td>
<td>9 (9.0)</td>
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<td>25 (51.0)</td>
<td>9 (18.4)</td>
<td>9 (18.4)</td>
<td>6 (12.2)</td>
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<td>4 (8.2)</td>
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<td>18 (36.7)</td>
<td>17 (34.7)</td>
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<tr>
<td>Domestic (^3)</td>
<td>19 (68.4)</td>
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<td>9 (47.4)</td>
<td>3 (15.8)</td>
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</tr>
<tr>
<td></td>
<td>13 (68.4)</td>
<td>12 (63.2)</td>
<td>2</td>
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<td></td>
</tr>
<tr>
<td>Type of meat</td>
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</tr>
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<td>30 (61.2)</td>
<td>17 (34.7)</td>
<td>7 (14.3)</td>
<td>5 (10.2)</td>
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<td>6 (12.2)</td>
</tr>
<tr>
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<td>30 (61.2)</td>
<td>26 (53.1)</td>
<td>9 (18.4)</td>
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</tr>
<tr>
<td>Breast</td>
<td>36 (52.8)</td>
<td>19 (52.8)</td>
<td>12 (33.3)</td>
<td>3 (8.3)</td>
<td>4 (11.1)</td>
</tr>
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<tr>
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<td>29 (80.6)</td>
<td>28 (77.8)</td>
<td>5 (13.9)</td>
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<td>Tenderloin</td>
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<td>3 (25.0)</td>
<td>2 (16.7)</td>
<td>1 (8.3)</td>
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<tr>
<td></td>
<td></td>
<td></td>
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<td></td>
<td>9 (75.0)</td>
</tr>
<tr>
<td>Others (^4)</td>
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<td>2 (66.7)</td>
<td>1 (33.3)</td>
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<tr>
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<td></td>
<td>2 (66.7)</td>
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<td></td>
<td></td>
<td>2 (66.7)</td>
</tr>
</tbody>
</table>

1): Other serotypes include Agona, Corvallis, Cerro, Duesseldorf, Typhimurium, and Muenchen
2): Parentheses show percentage
3): Location of a farm was not presented on a meat package
4): consisted of a mixed package of thigh and breast and a package without showing type of meat
Table 2. Antimicrobial resistance of *Salmonella enterica* subsp. *enterica* isolated from retail poultry meat

<table>
<thead>
<tr>
<th>Serotype</th>
<th>No. of isolates</th>
<th>ABPC</th>
<th>CTX</th>
<th>CP</th>
<th>FOM</th>
<th>GM</th>
<th>KM</th>
<th>SM</th>
<th>ST</th>
<th>TC</th>
<th>NA</th>
<th>CPFX</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>S</em>. Infantis</td>
<td>33</td>
<td>5 (15.2)</td>
<td>3 (9.1)</td>
<td>1 (3.0)</td>
<td>0</td>
<td>0</td>
<td>9 (27.3)</td>
<td>24 (72.7)</td>
<td>15 (45.5)</td>
<td>28 (84.8)</td>
<td>5 (15.2)</td>
<td>0</td>
</tr>
<tr>
<td><em>S</em>. Schwarzengrund</td>
<td>12</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>9 (75.0)</td>
<td>8 (66.7)</td>
<td>3 (25.0)</td>
<td>9 (75.0)</td>
<td>1 (8.3)</td>
<td>0</td>
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</tr>
<tr>
<td><em>S</em>. Manhattan</td>
<td>9</td>
<td>3 (33.3)</td>
<td>3 (33.3)</td>
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<td>0</td>
<td>0</td>
<td>8 (88.9)</td>
<td>0</td>
<td>8 (88.9)</td>
<td>1 (11.1)</td>
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<td><em>S</em>. Agona</td>
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<td>0</td>
<td>0</td>
<td>0</td>
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<td>0</td>
<td>1 (100)</td>
<td>0</td>
<td>0</td>
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</tr>
<tr>
<td><em>S</em>. Corvallis</td>
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<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
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<td></td>
</tr>
<tr>
<td><em>S</em>. Cerro</td>
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<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
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<td></td>
</tr>
<tr>
<td><em>S</em>. Duesseldorf</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>S</em>. Typhimurium</td>
<td>1</td>
<td>1 (100)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
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</tr>
<tr>
<td><em>S</em>. Muenchen</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>60</td>
<td>9 (15.0)</td>
<td>6 (10.0)</td>
<td>1 (1.7)</td>
<td>0</td>
<td>18 (30.0)</td>
<td>41 (68.3)</td>
<td>20 (33.3)</td>
<td>47 (78.3)</td>
<td>8 (13.3)</td>
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</tr>
</tbody>
</table>

ABPC, ampicillin; CTX, cefotaxime; CP, chloramphenicol; FOM, fosfomycin; GM, gentamicin; KM, kanamycin; SM, streptomycin; ST, sulfamethoxazole/trimethoprim; TC, tetracycline; NA, nalidixic acid; CPFX, ciprofloxacin.

Discs containing the following antibiotics were used for *Salmonella*: ampicillin (10 μg), cefotaxime (30 μg), chloramphenicol (30 μg), fosfomycin (50 μg), gentamicin (10 μg), kanamycin (30 μg), streptomycin (10 μg), sulfamethoxazole/trimethoprim (23.75/1.25 μg), tetracycline (30 μg), nalidixic acid (30 μg), and ciprofloxacin (5 μg).

1): Parentheses show percentages.
Table 3. Antimicrobial resistance of *Campylobacter jejuni* and *Campylobacter coli* isolated from retail poultry meat

<table>
<thead>
<tr>
<th>Species</th>
<th>No. of isolates</th>
<th>ABPC</th>
<th>CP</th>
<th>EM</th>
<th>KM</th>
<th>SM</th>
<th>TC</th>
<th>ST</th>
<th>NA</th>
<th>CPFX</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>C. jejuni</em></td>
<td>65</td>
<td>31 (47.7)</td>
<td>0</td>
<td>0</td>
<td>24 (36.9)</td>
<td>5 (7.7)</td>
<td>22 (33.8)</td>
<td>60 (92.3)</td>
<td>31 (47.7)</td>
<td>26 (40.0)</td>
</tr>
<tr>
<td><em>C. coli</em></td>
<td>9</td>
<td>3 (33.3)</td>
<td>0</td>
<td>0</td>
<td>3 (33.3)</td>
<td>1 (11.1)</td>
<td>2 (22.2)</td>
<td>7 (77.8)</td>
<td>4 (44.4)</td>
<td>4 (44.4)</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>74</td>
<td>34 (45.9)</td>
<td>0</td>
<td>0</td>
<td>27 (36.5)</td>
<td>6 (8.1)</td>
<td>24 (32.4)</td>
<td>67 (90.5)</td>
<td>35 (47.3)</td>
<td>30 (40.5)</td>
</tr>
</tbody>
</table>

ABPC, ampicillin; CP, chloramphenicol; EM, erythromycin; KM, kanamycin; SM, streptomycin; TC, tetracycline; ST, sulfamethoxazole/trimethoprim; NA, nalidixic acid; CPFX, ciprofloxacin

Discs containing the following antibiotics were used for *Campylobacter*: ampicillin (10 μg), chloramphenicol (30 μg), erythromycin (15 μg), kanamycin (30 μg), streptomycin (10 μg), tetracycline (30 μg), sulfamethoxazole/trimethoprim (23.75/1.25 μg), nalidixic acid (30 μg), and ciprofloxacin (5 μg).

1): Parentheses show percentages.
Fig. 1 c

Antibiogram

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<th>Source</th>
<th>Area of production</th>
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</tr>
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<td>KSAL-4476</td>
<td>breast</td>
<td>western</td>
</tr>
<tr>
<td>KSAL-4475</td>
<td>breast</td>
<td>western</td>
</tr>
<tr>
<td>KSAL-4376</td>
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<td>western</td>
</tr>
<tr>
<td>KSAL-4412</td>
<td>thigh</td>
<td>domestic</td>
</tr>
<tr>
<td>KSAL-4422</td>
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<td>western</td>
</tr>
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<td>KSAL-4408</td>
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</tr>
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<td>KSAL-4397</td>
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</table>
Fig. 2b