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Prevalence and Antimicrobial Resistance of *Salmonella* Recovered from Processed Poultry

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ABSTRACT

This study was conducted to determine the prevalence and antimicrobial resistance of *Salmonella* isolates recovered from processed poultry. Four hundred eighty pre- and postchill whole broiler chicken carcasses were collected from a poultry processing plant between July 2004 and June 2005. Water samples also were collected at the entrance and exit of the chiller. After preenrichment, carcass and water samples were analyzed for the presence of *Salmonella* using the automated BAX system followed by traditional culture methods. The proportions of pre- and postchill carcasses that were positive for *Salmonella* were 88.4 and 84.1%, respectively. Ninety-two percent of water samples collected at the entrance of the chiller were positive for *Salmonella,* but all exit samples were negative. There was no significant difference in the prevalence of *Salmonella* between pre- and postchill carcasses ($P > 0.05$). *Salmonella* isolates recovered were serotyped and tested for susceptibility to antimicrobials. Thirteen serotypes were identified; the most common were *Salmonella* Kentucky (59.5%) and *Salmonella* Typhimurium (17.8%). Three hundred thirty-nine (79.8%) of the isolates were resistant to at least one antimicrobial, and 53.4% were resistant to three or more antimicrobials. Resistance was most often observed to tetracycline (73.4% of isolates), ampicillin (52.9%), amoxicillin–clavulanic acid (52%), ceftiofur (51.7%), streptomycin (35.2%), and sulfisoxazole (21.8%). These results indicate the high prevalence of *Salmonella* contamination in whole broiler carcasses, and a large number of these *Salmonella* isolates were resistant to commonly used antimicrobials.

Salmonella is recognized as one of the major foodborne pathogens in the United States, causing an estimated 1.4 million cases of illness, approximately 20,000 hospitalizations, and more than 500 deaths annually *(25).* Although a growing number of human salmonellosis cases are associated with contaminated fruits and vegetables, traditionally illness has been linked with consumption of contaminated food of animal origin, especially poultry and poultry products *(4, 38, 45).* More problematic is the fact that antimicrobial resistance, in particular multidrug resistance (MDR), is being increasingly identified among numerous *Salmonella* serotypes recovered from animals and humans worldwide *(43, 47).* The levels and degree of resistance vary globally and are influenced by antimicrobial use practices in humans and animals and geographical variations in the epidemiology of *Salmonella* infections *(47).*

Salmonella isolates displaying resistance to clinically important antibiotics have been reported since the early 1960s, when most of the reported resistance was limited to a single antibiotic *(5, 9, 41).* However, since the mid-1970s, there has been an increasing trend of *Salmonella* isolates exhibiting MDR phenotypes worldwide. The recovery of antimicrobial-resistant *Salmonella* in foods of animal origin has raised concerns that the treatment of human salmonellosis may be compromised because antimicrobial-resistant strains appear to be more often associated with severe disease than are susceptible isolates *(16, 42).* Of significant concern is the isolation of *Salmonella* exhibiting decreased susceptibility to fluoroquinolones (e.g., ciprofloxacin) and extended-spectrum cephalosporins (e.g., ceftiofur and ceftriaxone), because these two antimicrobial classes are important in treating *Salmonella* infections in adults and children, respectively *(13, 26, 47).*

The majority of these antimicrobial-resistant phenotypes in *Salmonella* and other pathogens are gained from extrachromosomal genes that may impart resistance to an entire antimicrobial class. In recent years, a number of these resistance genes have been associated with large transferable plasmids on which may be other DNA mobile elements, such as transposons and integrons. Recent data indicate that different resistance determinants can amass in linked clusters, such that antimicrobials of a different class or substances such as disinfectants or heavy metals may select for MDR in bacteria *(14, 15).* Although resistance, in particular MDR, appears to be most serious in certain serotypes, this situation may be shifting. Thus, there is a continuing need for increased surveillance of antimicrobial-resistant phenotypes in *Salmonella* isolates of animal and human origin on a global basis.

The role of meat and poultry products in the dissemination of antimicrobial-resistant zoonotic bacterial pathogens also is well documented *(23, 43, 44, 47).* Although the hygienic standards for meat production are quite high

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in most developed countries, fecal contamination of meat products cannot be completely prevented. For example, Izat et al. *(17)* reported that chill water and the chilling process can serve as an important source of pathogen contamination between carcasses. As a result, a small number of contaminated carcasses may have a large impact on contamination. Other procedures such as handling during processing also may contribute to cross-contamination among carcasses *(3).* Recently, several investigators suggested that processing conditions may play a significant role in promoting and influencing the selection of pathogens, including antimicrobial-resistant variants *(21, 23, 29, 32).* However, the factors that contribute to this selection have yet to be fully evaluated.

Several studies have been conducted on the prevalence and antimicrobial resistance of *Salmonella* in processed poultry, poultry products, and poultry processing plants *(18, 23, 27, 32, 34).* However, little information is available about the prevalence and antimicrobial resistance of *Salmonella* in pre- and postchill whole broiler carcasses from U.S. mid-Atlantic processing plants. The objectives of this study were (i) to determine the prevalence of *Salmonella* in pre- and postchill carcasses; (ii) to serotype *Salmonella* isolates from pre- and postchill carcasses, and (iii) to determine antimicrobial resistance profiles for these isolates.

MATERIALS AND METHODS

Sample collection. Whole broiler carcass samples were collected at monthly intervals between July 2004 and June 2005 from a processing plant in the mid-Atlantic region. Prechill samples were obtained after the last visual inspection just before the carcass was loaded into the chill tank. Postchill samples were obtained immediately after removal of the carcass from the chill tank as carcasses were being rehung onto the line for further processing. Samples were collected from a total of 20 prechill carcasses and 20 postchill carcasses during each visit. On a given sampling day, the carcasses originated from a single producer or farm. A water sample also was collected at the entrance where the carcasses entered the chiller and at the exit where the carcasses left the chiller. The water temperature at the chiller entrance and exit was monitored with a thermometer (Fisher Scientific, Boston, Mass.). Total chlorine, free chlorine, and pH were measured with a HACH test kit pocket colorimeter II (HACH Company, Loveland, Colo.). Each carcass was placed in a 3,500-ml sterile plastic stomacher bag (Fisher) following aseptic techniques in the processing plant. All samples were placed in coolers with ice and transported to the laboratory within 1 h of collection and were processed immediately.

Microbiological analysis. Five hundred milliliters of sterile buffered peptone water (BPW; Fisher) was added to the interior and exterior surfaces of each carcass, and the carcass in the bag was vigorously shaken for 1 min. The bag containing the whole carcass and rinse solution was incubated at 37°C for 24 h *(36).* After incubation, the samples were screened for *Salmonella* using the BAX system, a commercial PCR-based system (DuPont Qualicon Inc., Wilmington, Del.). Samples positive for *Salmonella* with the BAX system were added to two enrichment broths, 0.5 ml into tetrathionate (Difco, Becton Dickinson, Detroit, Mich.) (Hajna) broth tubes and 0.1 ml into Rappaport Vassiliadis (Difco, Becton Dickinson) broth tubes, and tubes were incubated at 42°C for 24 h. Enrichment solutions were streaked onto xylose lysine agar supplemented with Tergitol 4 (XLT4; Difco, Becton Dickinson) and brilliant green sulfa agar (BGSA; Difco, Becton Dickinson) containing 10 ppm novobiocin (Sigma-Aldrich, Atlanta, Ga.), and the plates were incubated at 37°C for 24 h. Typical colonies (maximum of five) were randomly selected from each plate and streaked onto tryptic soy agar (Difco, Becton Dickinson) for purification; these plates were incubated at 37°C for 24 h. Suspected *Salmonella* colonies were inoculated onto slant tubes of lysine iron agar (Difco, Becton Dickinson) and triple sugar iron agar (Difco, Becton Dickinson) and incubated at 37°C for 24 h. Presumptive colonies were confirmed *(36)* using polyvalent serum A-Vi (Becton Dickinson, Sparks, Md.) by a slide agglutination test according to the manufacturer's instructions. To detect *Salmonella* in water samples, 10 ml of each water sample was added to 10 ml of double-strength BPW (Difco, Becton Dickinson), incubated at 37°C for 24 h, and confirmed as described above. *Salmonella* isolates were stored at −72°C in tryptic soy broth (Difco, Becton Dickinson) with 15% glycerol. One isolate from each positive sample was chosen from the XLT4 plates for further serotyping and antimicrobial susceptibility testing. If no colony could be verified as *Salmonella,* one isolate from the corresponding BGSA plate was used for further serotyping and antimicrobial susceptibility testing. Overall, 91 and 9% of isolates were chosen from XLT4 and BGSA plates, respectively.

Serotyping of *Salmonella.* A total of 425 *Salmonella* isolates were serotyped by standard methods using reagents prepared in accordance with World Health Organization guidelines at the *Salmonella* Reference Center (School of Veterinary Medicine, University of Pennsylvania, Kennett Square).

Antimicrobial susceptibility testing. Antimicrobial MICs were determined using the Sensititre automated antimicrobial susceptibility system in accordance with the manufacturer's instructions (Trek Diagnostic Systems, Westlake, Ohio) at the *Salmonella* Reference Center. Results were interpreted in accordance with testing standards and interpretive criteria provided by Clinical and Laboratory Standards Institute *(10, 11).*

The following MIC ranges were tested for 15 antimicrobials of veterinary and human health importance: amikacin (0.5 to 32 μg/ml), amoxicillin–clavulanic acid (0.5/1 to 16/32 μg/ml), ampicillin (1 to 32 μg/ml), cefoxitin (0.5 to 32 μg/ml), ceftiofur (0.12 to 8 μg/ml), ceftriaxone (0.5 to 64 μg/ml), chloramphenicol (2 to 32 μg/ml), ciprofloxacin (0.015 to 4 μg/ml), gentamicin (0.25 to 16 μg/ml), kanamycin (8 to 64 μg/ml), nalidixic acid (0.5 to 32 μg/ml), streptomycin (32 to 64 μg/ml), sulfisoxazole (16 to 512 μg/ml), tetracycline (4 to 32 μg/ml), and trimethoprim-sulfamethoxazole (0.12/2.38 to 4/76 μg/ml). *Escherichia coli* ATCC 25922, *E. coli* ATCC 35218, *Enterococcus faecalis* ATCC 51299, and *Pseudomonas aeruginosa* ATCC 27853 were used as controls.

Statistical analysis. Differences in the prevalence of *Salmonella* isolated from pre- and postchill carcasses were determined with an analysis of variance (ANOVA, $P < 0.05$) using a randomized complete block design with month as the blocking term. This method also was used to determine the significance of differences among antimicrobial resistance phenotypes observed in *Salmonella* isolates from pre- and postchill carcasses. Percentage data were transformed prior to the ANOVA using the arcsine transformation *(22).* All computations were performed with the STATISTIX 8.0 software program (Analytical Software, Tallahassee, Fla.).

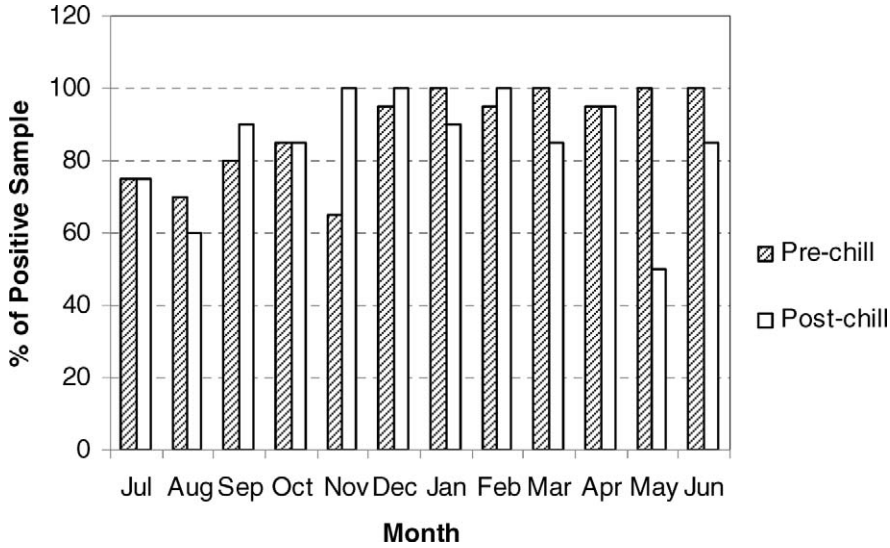

FIGURE 1. *Prevalence of* Salmonella *on pre- and postchill broiler carcasses from July 2004 to June 2005.*

RESULTS AND DISCUSSION

Prevalence of *Salmonella.* A total of 480 whole broiler carcasses were obtained from two selected points on the processing line (prechill and postchill), and 24 water samples were collected at the entrance and exit of the chiller from July 2004 to June 2005. Samples were analyzed for the presence of *Salmonella* using the automated BAX system and then traditional culture methods. The proportions of prechill and postchill carcass samples that were positive for *Salmonella* were 88.4 and 84.1%, respectively. All water samples collected at the entrance of the chiller were positive for *Salmonella* except for those collected in the month of October. In contrast, no water samples obtained at the chiller exit were positive for *Salmonella.*

Although *Salmonella* contamination rates of tested carcass samples were both above 80%, no significant difference ($P > 0.05$) was observed in the prevalence of *Salmonella* on pre- and postchill carcasses. A higher prevalence of *Salmonella* was observed in samples from postchill than those from prechill carcasses with the exception of those samples collected in August, January, March, May, and June. However, no seasonal effects with regards to *Salmonella* prevalence on broiler carcasses (either pre- or postchill) were observed over the 1-year study period (Fig. 1). In contrast, Logue et al. *(23)* found that of 2,411 turkey carcass samples tested at a Midwestern poultry plant, only 402 (16.7%) were positive for *Salmonella* and that *Salmonella* was more frequently recovered from prechill than from postchill carcasses. Logue et al. *(23)* also reported a seasonal effect in the incidence of *Salmonella* on freshly processed turkey; they recovered *Salmonella* more frequently from carcasses in the spring and summer months than in the autumn and winter months. Differences between our results and those of Logue et al. *(23)* may be due to several factors, including variation in sample location (mid-Atlantic versus Midwest), type of poultry (chicken versus turkey), sampling procedures (whole carcass rinse versus whole carcass enrichment), poultry contamination levels, postprocessing contamination, differences in sample size (480 versus 2,411), and *Salmonella* detection methods (BAX system versus immunomagnetic separation). In particular, the whole carcass method for recovery of *Salmonella* has been shown to be more sensitive than the swab or carcass rinse method *(35)* because it can recover *Salmonella* cells that are firmly attached, entrapped, or loosely attached to the surface or the inside of a carcass *(20, 36).*

Temperature, pH, free chlorine, and total chlorine in the water at the entrance and exit of the chiller were recorded every month. The average water temperature was 17°C at the entrance and 3.9°C at the exit. The average pHs of the chiller water at the entrance and exit were 8.6 and 7.4, respectively, and average concentrations of free and total chlorine in chiller water were 7.5 and 9.4 ppm, respectively. There was no correlation between the presence of *Salmonella* on postchill carcasses and the concentrations of free and total chlorine in the chiller water, indicating that the chilling process did not have a major effect on *Salmonella* prevalence. However, further research is needed to determine the effectiveness of these mitigation steps, because previous reports have differed in their interpretation of whether the chilling and chlorination processes significantly reduce pathogen levels on poultry carcasses or potentially select for particular pathogens, i.e., cold- and chlorine-resistant variants *(21, 23, 29, 32).*

Distribution of *Salmonella* serotypes. Four hundred ten (96.5%) of the total 425 *Salmonella* isolates from prechill, postchill, and water samples were typeable with standard antisera. Thirteen different *Salmonella* serotypes were identified; the most frequent were Kentucky (59.5%), Typhimurium (17.8%), Litchfield (5.8%), and Mbandaka (5.4%) (Table 1). *Salmonella* Kentucky accounted for 51.4, 67.3, 72.7% of prechill, postchill, and water sample isolates, respectively. Other *Salmonella* serotypes commonly recovered from prechill and postchill samples were Typhimurium (20.7 and 15.3% of samples, respectively), Litchfield (8.4 and 3.4%), Mbandaka (4.7 and 5.9%), and Mono group C (3.7 and 0.9%). *Salmonella* Typhimurium (9.1%), *Salmonella* Mbandaka (9.1%), and *Salmonella* Mono group C (9.1%) also were recovered from water samples in ad-

TABLE 1. *Distribution of* Salmonella *serotypes recovered from prechill and postchill broiler carcass and water samples*

Salmonella serotype	% of isolates in samples from: Prechill (n = 212)	Postchill (n = 202)	Water (n = 11)	Total (n = 425)
Kentucky	51.4	67.3	72.7	59.5
Typhimurium	20.7	15.3	9.1	17.8
Litchfield	8.4	3.4	0	5.8
Mbandaka	4.7	5.9	9.1	5.4
Schwarzengrund	2.3	2.4	0	2.3
Rough	4.2	2.9	0	3.5
Mono group C	3.7	0.9	9.1	2.5
Senftenberg	0.9	0	0	0.5
Unnamed group B	1.4	0.9	0	1.2
Heidelberg	0.5	0	0	0.2
Thompson	0.5	0	0	0.2
Virchow	0.5	0	0	0.2
Worthington	0.5	0	0	0.2
Newington	0	0.5	0	0.2

dition to *Salmonella* Kentucky (Table 1). *Salmonella* serotypes rarely recovered were Senftenberg, Thompson, Newington, Heidelberg, Virchow, and Worthington (Table 1). Several *Salmonella* serotypes were repeatedly recovered from sampling during the year, which is consistent with the results of Nde et al. *(28),* who reported the recovery of similar serotypes on different visits to a commercial turkey processing plant, suggesting that cross-contamination occurred during processing. Our data are similar to recent U.S. Department of Agriculture (USDA) Food Safety and Inspection Service broiler data for 2004 to 2005, which indicated that the most common *Salmonella* serotypes recovered were Kentucky, Typhimurium, Mbandaka, Schwarzengrund, Heidelberg, and Thompson *(39).* The association of specific serotypes with poultry might be related to host adaptation or other unknown factors *(40).* Of the 13 *Salmonella* serotypes recovered in our study, several serotypes, including Typhimurium, Mbandaka, Heidelberg, and Thompson, were also among the top 20 serotypes that cause human salmonellosis in the United States *(8, 30).* The *Salmonella* serotypes identified in our study were also consistent with those reported in similar studies *(6, 33).* For example, both Byrd et al. *(6)* and Roy et al. *(33)* reported that *Salmonella* Kentucky was one of the predominant serotypes recovered from either broiler hatcheries and farms or poultry and poultry products, respectively. In contrast, Bailey et al. *(1)* found that *Salmonella* Thompson and *Salmonella* Molade were the predominant serotypes in broilers and in broiler hatchery and processing environments, whereas Logue et al. *(23)* reported that *Salmonella* serotypes Agona, Hadar, Heidelberg, and Senftenberg were the most common among the isolates recovered from pre- and postchill turkey carcasses. Logue et al. *(23)* also recovered similar serotypes from pre- and postchill carcasses, which is consistent with the results of the present study.

Antimicrobial resistance phenotypes. *Salmonella* isolates were tested for susceptibility to 15 antimicrobial agents of veterinary and human health significance. Three hundred thirty-nine (79.8%) isolates were resistant to at least one antimicrobial, and 53.4% were resistant to three or more antimicrobials. Overall, the most common resistance phenotypes were those to tetracycline (73.4%), ampicillin (52.9%), amoxicillin–clavulanic acid (52%), cefoxitin (52%), and ceftiofur (51.7%) (Table 2). Less resistance was found to streptomycin (35.2%), sulfisoxazole (21.8%), and kanamycin (6.3%). All isolates were uniformly susceptible to amikacin, ceftriaxone, ciprofloxacin, chloramphenicol, and nalidixic acid (Table 2). The high rate of tetracycline resistance in our study is not surprising because tetracyclines are some of the most commonly used antimicrobials in food animal production and resistance phenotypes to these compounds are frequently observed in *Salmonella* isolates worldwide *(7, 23, 24, 37, 43).*

TABLE 2. *Antimicrobial resistance phenotypes of* Salmonella *isolates recovered from prechill and postchill carcass and water samples*

Antibiotic	Resistant breakpoint (μg/ml)[a]	% of resistant isolates in samples from: Prechill (n = 212)	Postchill (n = 202)	Water (n = 11)	Total (n = 425)
Tetracycline	32	69.3	78.7	54.5	73.4
Ampicillin	32	57.0	48.5	54.5	52.9
Amoxicillin–clavulanic acid	16/32	56.6	47.5	45.4	52
Cefoxitin	32	57.0	47.0	45.4	52
Ceftiofur	8	56.1	47.5	45.4	51.7
Streptomycin	64	31.1	40.0	27.2	35.2
Sulfisoxazole	512	26.4	17.3	18.1	21.8
Kanamycin	64	8.4	4.4	0	6.3
Gentamicin	16	1.4	0	0	0.7
Ceftriaxone	64	0	0	0	0
Amikacin	32	0	0	0	0
Nalidixic acid	32	0	0	0	0
Chloramphenicol	32	0	0	0	0
Ciprofloxacin	4	0	0	0	0
Trimethoprim-sulfamethoxazole	4/76	0	0	0	0

[a] MICs were determined via microdilution broth methods in accordance with CLSI standards *(10, 11).*

The most common resistance phenotypes observed among prechill and postchill *Salmonella* isolates were to tetracycline (69.3 and 78.7% of isolates, respectively), ampicillin (57 and 48.5%), amoxicillin–clavulanic acid (56.6 and 47.5%), cefoxitin (57 and 47%), ceftiofur (56.1 and 47.5%), streptomycin (31.1 and 40%), and sulfisoxazole (26.4 and 17.3%) (Table 2). There were no significant differences in the incidence of antimicrobial-resistant *Salmonella* isolates from pre- and postchill carcasses ($P > 0.05$), suggesting that chilling has no apparent selection effect on *Salmonella* antimicrobial resistance phenotypes. Similar antimicrobial resistance phenotypes were also observed among *Salmonella* isolates recovered from water samples.

TABLE 3. *Antimicrobial resistance profiles of* Salmonella *isolates recovered from prechill and postchill carcass and water samples*

Resistance or susceptibility profile[a]	% of isolates in samples from: Prechill (n = 212)	Postchill (n = 202)	Water (n = 11)
T-A-Am-C-Ce-Su-K	7.5	4.0	0
T-A-Am-C-Ce-St	22.2	19.8	18.2
T-A-Am-C-Ce-Su	11.3	5.9	0
T-A-Am-C-Ce	7.5	12.3	0
A-Am-C-Ce	7.0	4.5	18.2
T-St	8.1	18.3	0
T-Su	5.2	6.4	9.1
T	4.2	9.4	18.2
T-Su-G	1.4	0	0
T-A	0	1.0	0
Other[b]	1.4	19.8	18.2
Susceptible to all tested antimicrobials	24.0	16.3	18.2

[a] T, tetracycline; A, ampicillin; Am, amoxicillin; C, cefoxitin; Ce, ceftiofur; Su, sulfisoxazole; K, kanamycin; St, streptomycin; G, gentamicin.

[b] These resistance profiles consisted of three profiles from prechill carcass, four profiles from postchill carcass, and two profiles from water samples (not shown). Each profile was represented by only one isolate.

These findings are comparable to data from other studies *(7, 12, 24)* in which high antimicrobial resistance rates were found among *Salmonella* isolates recovered from poultry or poultry products.

MDR (resistance to two or more antimicrobials) was observed in 72.5% of the *Salmonella* isolates recovered. Most of the *Salmonella* isolates (45.8%) displayed resistance to an average of five or more tested antimicrobials, a finding similar to that reported by Logue et al. *(23).* Two isolates were resistant to eight different antimicrobials (data not shown). Twelve, 13, and 6 antimicrobial resistance profiles were observed among *Salmonella* isolates recovered from prechill and postchill carcass and water samples, respectively. The predominant MDR profiles for pre- and postchill isolates were tetracycline–ampicillin–amoxicillin–clavulanic acid–cefoxitin–ceftiofur–streptomycin (T-A-Am-C-Ce-St; 22.2 and 19.8% of isolates, respectively), tetracycline–ampicillin–amoxicillin–clavulanic acid–cefoxitin–ceftiofur–sulfisoxazole (T-A-Am-C-Ce-Su; 11.3 and 5.9%), and tetracycline-streptomycin (T-St; 8.1 and 18.3%) (Table 3). Our results show that a large number of recovered *Salmonella* isolates were resistant to multiple antimicrobials. Multidrug-resistant *Salmonella* isolates have been reported as the cause of both human and animal salmonellosis worldwide by numerous investigators, and these strains are of particular clinical concern because they frequently display resistance to key antimicrobials, notably third-generation cephalosporins *(18, 23, 43, 46).*

Association of *Salmonella* serotype and antimicrobial resistance phenotype. There did not appear to be an association between antimicrobial resistance phenotype and a particular serotype; however, several notable exceptions were observed. For example, the majority of *Salmonella* Kentucky and *Salmonella* Typhimurium isolates displayed resistance to tetracycline and beta-lactam antimicrobials (e.g., ampicillin and amoxicillin–clavulanic acid), but much lower rates of resistance were found among other serotypes including Litchfield, Mbandaka, and Schwarzengrund (Table 4). Similar findings of differences in antimicrobial resistance profiles among *Salmonella* serotypes have been reported by other investigators *(2, 7, 19, 27, 31, 46).*

TABLE 4. *Antimicrobial resistance phenotypes of* Salmonella *serotypes*[a]

Antibiotic	% of resistant isolates of serotypes: Ken (n = 253)	Typ (n = 76)	Lit (n = 25)	Mba (n = 23)	Rou (n = 15)	GrC (n = 11)	Sch (n = 10)	GrB (n = 5)	Hei (n = 1)
Tetracycline	86.2	89.5	4	17.3	93.3	36.4	0	60	0
Ampicillin	62.8	61.8	0	4.3	73.3	27.3	10	40	100
Amoxicillin–clavulanic acid	61.3	61.8	0	4.3	73.3	27.3	10	40	100
Cefoxitin	61.3	61.8	0	4.3	73.3	27.3	10	40	100
Ceftiofur	61.3	61.8	0	4.3	66.7	27.3	10	40	100
Streptomycin	55.3	7.9	0	4.3	13.3	0	0	20	0
Sulfisoxazole	3.9	82.8	4	13	66.7	36.4	0	40	0
Kanamycin	1.6	25	0	0	6.7	18.2	0	20	0
Gentamicin	0	3.9	0	0	0	0	0	0	0
Ceftriaxone	0	0	0	0	0	0	0	0	0
Amikacin	0	0	0	0	0	0	0	0	0
Nalidixic acid	0	0	0	0	0	0	0	0	0
Chloramphenicol	0	0	0	0	0	0	0	0	0
Ciprofloxacin	0	0	0	0	0	0	0	0	0
Trimethoprim-sulfamethoxazole	0	0	0	0	0	0	0	0	0

[a] Ken, Kentucky; Typ, Typhimurium; Lit, Litchfield; Mba, Mbandaka; Rou, Rough; GrC, Group C; Sch, Schwarzengrund; GrB, Group B; Hei, Heidelberg. *Salmonella* serotypes Senftenberg, Thompson, Virchow, Worthington, and Newington were susceptible to all tested antimicrobials. (Antimicrobial resistant breakpoint concentrations are provided in Table 2.)

TABLE 5. *Antimicrobial resistance profiles of* Salmonella *serotypes*[a]

Resistance or susceptibility profile[b]	% of isolates of serotypes: Ken (n = 253)	Typ (n = 76)	Lit (n = 25)	Mba (n = 23)	Rou (n = 15)	Sch (n = 10)	GrC (n = 11)	GrB (n = 5)	Hei (n = 1)
T-A-Am-C-Ce-Su-K	1.2	23.7	0	0	0	0	18.2	20	0
T-A-Am-C-Ce-St	32.8	3.9	0	4.3	13.3	0	0	0	0
T-A-Am-C-Ce-Su	1.2	31.6	0	0	46.7	0	9.1	20	0
T-A-Am-C-Ce	15.4	1.3	0	0	6.7	0	0	0	0
A-Am-C-Ce	9.5	0	0	0	0	10	0	0	100
T-St	20.6	1.3	0	0	0	0	0	20	0
T-Su	0.8	21.1	4	13.0	13.3	0	9.1	0	0
T	11.5	0	0	0	6.7	0	0	0	0
T-Su-G	0	3.9	0	0	0	0	0	0	0
T-A	0.8	0	0	0	0	0	0	0	0
Other[c]	2.8	1.3	6.7	0	0	0	0	0	0
Susceptible to all tested antimicrobials	3.6	11.8	96	82.6	6.7	90	63.4	40	0

[a] Ken, Kentucky; Typ, Typhimurium; Lit, Litchfield; Mba, Mbandaka; Rou, Rough; GrC, Group C; Sch, Schwarzengrund; GrB, Group B; Hei, Heidelberg.

[b] T, tetracycline; A, ampicillin; Am, amoxicillin; C, cefoxitin; Ce, ceftiofur; Su, sulfisoxazole; K, kanamycin; St, streptomycin; G, gentamicin.

[c] These resistance profiles consisted of seven profiles from *Salmonella* Kentucky, one profile from *Salmonella* Typhimurium, and one profile from *Salmonella* Rough. Each profile was represented by only one isolate. *Salmonella* serotypes Senftenberg, Thompson, Virchow, Worthington, and Newington were susceptible to all tested antimicrobials.

In our study, 89.5% of *Salmonella* Typhimurium and 84.2% of *Salmonella* Kentucky isolates were MDR (data not shown). The predominant MDR profiles for *Salmonella* Kentucky isolates were T-A-Am-C-Ce-St (32.8% of isolates), T-St (20.6%), and T-A-Am-C-Ce (15.4%), and the major MDR profiles observed among *Salmonella* Typhimurium isolates were T-A-Am-C-Ce-Su (31.6%), T-A-Am-C-Ce-Su-K (23.7%), and T-Su (21.1%) (Table 5). The notable difference in MDR profiles between these two serotypes was the association of streptomycin resistance more often with *Salmonella* Kentucky isolates as compared with sulfisoxazole resistance, which was more often associated with *Salmonella* Typhimurium isolates. This difference is most likely due to differences among isolates of these two serotypes with regard to possession of large transferable plasmids on which other mobile DNA elements, such as transposons and integrons, may be found. These mobile DNA elements can transmit genetic determinants for several different antimicrobial resistance mechanisms and may account for the rapid dissemination of resistance genes among different bacteria *(14, 47).*

The results of the study presented here demonstrate a high prevalence of *Salmonella* contamination and varied spectrum of antimicrobial resistance, including several MDR phenotypes, among *Salmonella* isolates from pre- and postchill whole broiler carcass samples. Overall, antimicrobial resistance phenotypes were similar between *Salmonella* isolates recovered from pre- and postchill samples. These data emphasize the public health importance of continuing efforts to educate consumers in proper food hygiene and highlight the need for continued surveillance of zoonotic foodborne pathogens, including antimicrobial-resistant variants, throughout the food production continuum.

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REFERENCES

1. Bailey, J. S., N. J. Stern, P. Fedorka-Cray, S. E. Craven, N. A. Cox, D. E. Cosby, S. Ladely, and M. T. Musgrove. 2001. Sources and movement of *Salmonella* through integrated poultry operations: a multistate epidemiological investigation. *J. Food Prot.* 64:1690–1697.
2. Beach, J. C., E. A. Murano, and G. R. Acuff. 2002. Serotyping and antibiotic resistance profiling of *Salmonella* in feedlot and nonfeedlot beef cattle. *J. Food Prot.* 65:1694–1699.
3. Beery, J. T., M. B. Hugdahl, and M. P. Doyle. 1988. Colonization of gastrointestinal tract of chicks by *Campylobacter jejuni. Appl. Environ. Microbiol.* 54:2365–2370.
4. Bryan, F. L., and M. P. Doyle. 1995. Health risks and consequences of *Salmonella* and *Campylobacter jejuni* in raw poultry. *J. Food Prot.* 61:326–344.
5. Bulling, E., R. Stephan, and V. Sebek. 1973. The development of antibiotic resistance among *Salmonella* bacteria of animal origin in the Federal Republic of Germany and West Berlin. First communication: a comparison between the years of 1961 and 1970–1971. *Zentbl. Bakteriol.* 225:245–256.
6. Byrd, J. A., J. R. Deloach, D. E. Corrier, D. J. Nisbet, and L. H. Starker. 1999. Evaluation of *Salmonella* serotype distribution from commercial broiler hatcheries and grower houses. *Avian Dis.* 43:39–47.
7. Carraminana, J. J., C. Rota, U. Agustin, and A. Herrera. 2004. High prevalence of multiple resistance to antibiotics in *Salmonella* serovars isolated from a poultry slaughterhouse in Spain. *Vet. Microbiol.* 104:133–139.
8. Centers for Disease Control and Prevention. 2004. *Salmonella* surveillance: annual summary, 2003. Centers for Disease Control and Prevention, Atlanta.

9. Cherubin, C. E. 1981. Antibiotic resistance of *Salmonella* in Europe and the United States. *Rev. Infect. Dis.* 6:1105–1125.
10. Clinical and Laboratory Standards Institute. 2002. Performance standards for antimicrobial disk and dilution susceptibility tests for bacteria isolated from animals. Approved standard. CLSI/NCCLS document M31-A2. CLSI, Wayne, Pa.
11. Clinical and Laboratory Standards Institute. 2006. Performance standards for antimicrobial susceptibility testing. Sixteenth informational supplement. CLSI/NCCLS document M100-S16. CLSI, Wayne, Pa.
12. Duffy, G., O. M. Clock, M. G. O'Sullivan, A. Guillet, J. J. Sheridan, I. S. Blair, and D. A. McDowell. 1999. The incidence and antibiotic resistance profiles of *Salmonella* spp. on Irish retail meat products. *Food Microbiol.* 16:623–631.
13. Gupta, A., J. Fontana, C. Crowe, B. Bolstorff, A. Stout, S. Van Duyne, M. P. Hoekstra, J. M. Whichard, T. J. Barrett, and F. J. Angulo. 2003. Emergence of multidrug-resistant *Salmonella enterica* serotype Newport infections resistant to expanded-spectrum cephalosporins in the United States. *J. Infect. Dis.* 188:1707–1716.
14. Hall, R. M., C. M. Collis, M. J. Kim, S. R. Partridge, G. D. Recchia, and H. W. Stokes. 1999. Mobile gene cassettes and integrons in evolution. *Ann. N.Y. Acad. Sci.* 870:68–80.
15. Harbottle, H., S. Thakur, S. Zhao, and D. G. White. 2006. Genetics of antimicrobial resistance. *Anim. Biotechnol.* 17:111–124.
16. Helms, M., P. Vastrup, P. Gerner-Smidt, and K. Molbak. 2002. Excess mortality associated with antimicrobial drug-resistant *Salmonella* Typhimurium. *Emerg. Infect. Dis.* 8:490–495.
17. Izat, A. L., C. D. Driggers, M. Colberg, M. A. Reiber, and M. H. Adam. 1989. Comparison of the DNA probe to culture methods for the detection of *Salmonella* on poultry carcasses and processing waters. *J. Food Prot.* 52:564–570.
18. Jain, S., and J. Chen. 2006. Antibiotic resistance profiles and cell surface components of salmonellae. *J. Food Prot.* 69:1017–1023.
19. Larkin, C., C. Poppe, B. McNab, B. McEwen, A. Mahdi, and J. Odumeru. 2004. Antibiotic resistance of *Salmonella* isolated from hog, beef, and chicken carcass samples from provincially inspected abattoirs in Ontario. *J. Food Prot.* 67:448–455.
20. Lillard, H. S. 1988. Comparison of sampling methods and implications for bacterial decontamination of poultry carcasses by rinsing. *J. Food. Prot.* 51:405–408.
21. Lisle, J. T., S. C. Broadway, A. M. Prescott, B. H. Pyle, C. Fricker, and G. A. McFeters. 1998. Effects of starvation on physiological activity and chlorine disinfection in *Escherichia coli* O157:H7. *Appl. Environ. Microbiol.* 64:4658–4662.
22. Little, T. M., and F. J. Hills. 1978. Agricultural experimentation: design and analysis. John Wiley and Sons, New York.
23. Logue, C. M., J. S. Sherwood, P. A. Olah, L. M. Elijah, and M. R. Dockter. 2003. The incidence of antimicrobial-resistant *Salmonella* spp. on freshly processed poultry from US Midwestern processing plants. *J. Appl. Microbiol.* 94:16–24.
24. Manie, T., S. Khan, V. S. Brozel, W. J. Veith, and P. A. Gouws. 1998. Antimicrobial resistance of bacteria isolated from slaughtered retail chicken in South Africa. *Lett. Appl. Microbiol.* 26:253–258.
25. Mead, P. S., L. Slutsker, V. Dietz, L. F. McCaig, J. S. Bresee, C. Chapiro, P. M. Griffin, and R. V. Tauxe. 1999. Food-related illness and death in the United States. *Emerg. Infect. Dis.* 5:607–625.
26. Molbak, K., D. L. Baggesen, F. M. Aarestrup, J. M. Ebbesen, J. Engberg, K. Frydendahl, P. Gerner-Smidt, A. M. Petersen, and H. C. Wegener. 1999. An outbreak of multidrug-resistant, quinolone-resistant *Salmonella enterica* serotype Typhimurium DT104. *N. Engl. J. Med.* 341:1420–1425.
27. Nayak, R., and P. B. Kenney. 2002. Screening of *Salmonella* isolates from a turkey production facility for antibiotic resistance. *Poult. Sci.* 81:1496–1500.
28. Nde, C. W., J. S. Sherwood, C. Doetkott, and C. M. Logue. 2006. Prevalence and molecular profiles of *Salmonella* collected at a commercial turkey processing plant. *J. Food Prot.* 69:1794–1801.
29. Norwood, D. E., and A. Gilmour. 2000. The growth and resistance to sodium hypochlorite of *Listeria monocytogenes* in a steady-state multispecies biofilm. *J. Appl. Microbiol.* 88:512–520.
30. Olsen, S. J., R. Bishop, F. W. Brenner, T. H. Roels, N. Bean, R. V. Tauxe, and L. Slutsker. 2001. The changing epidemiology of *Salmonella*: trends in serotypes isolated from humans in the United States, 1987–1997. *J. Infect. Dis.* 183:753–761.
31. Poppe, C., G. Ayroud, G. Ollis, M. Chirino-Trejo, N. Smart, S. Quessy, and P. Micheal. 2001. Trends in antimicrobial resistance of *Salmonella* isolated from animals, food of animal origin, and the environment of animal production in Canada. *Microbiol. Drug Resist.* 7:197–212.
32. Rowe, M. T., G. Dunstall, R. Kirk, C. F. Loughney, J. L. Cooke, and S. R. Brown. 1998. Development of an image system for the study of viable but non-culturable forms of *Campylobacter jejuni* and its use to determine their resistance to disinfectants. *Int. J. Food Microbiol.* 15:491–498.
33. Roy, P., A. S. Dhillon, L. H. Lauerman, D. M. Schaberg, D. Bandli, and S. Johnson. 2002. Results of *Salmonella* isolation from poultry products, poultry environment, and other characteristics. *Avian Dis.* 46:17–24.
34. Sánchez, M. X., W. M. Fluckey, M. M. Brashears, and S. R. McKee. 2002. Microbial profile and antibiotic susceptibility of *Campylobacter* spp. and S*almonella* spp. in broilers processed in air-chilled and immersion-chilled environments. *J. Food Prot.* 65:948–956.
35. Simmons, M., D. L. Fletcher, M. E. Berrang, and J. A. Cason. 2003. Comparison of sampling methods for the detection of *Salmonella* on whole broiler carcasses purchased from retail outlets. *J. Food Prot.* 66:1768–1770.
36. Simmons, M., D. L. Fletcher, J. A. Cason, and M. E. Berrang. 2003. Recovery of *Salmonella* from retail broilers by a whole-carcass enrichment procedure. *J. Food Prot.* 66:446–450.
37. Threlfall, E. J., L. R. Ward, and B. Rowe. 1997. Increasing incidence of resistance to trimethoprim and ciprofloxacin in epidemic *Salmonella* Typhimurium DT104 in England and Wales. *Eur. Rep.* 2:11–16.
38. Todd, E. C. D. 1980. Poultry-associated foodborne disease—its occurrence, cost, sources, and prevention. *J. Food Prot.* 43:129–139.
39. U.S. Department of Agriculture, Food Safety and Inspection Service. 2006. Serotype profile of *Salmonella* isolates from meat and poultry products January 1998 through December 2005. Available at: http://www.fsis.usda.gov/Science/Serotypes_Profile_Salmonella_Isolates/index.asp. Accessed 15 December 2006.
40. Uzzau, S., D. J. Brown, T. Wallis, S. Rubino, G. Leori, S. Bernard, J. Casadesus, D. J. Platt, and J. E. Olsen. 2000. Host-adapted serotypes of *Salmonella enterica. Epidemiol. Infect.* 125:229–255.
41. Van Leeuwen, W. J., J. Van Embden, P. Guinee, E. H. Kampelmacher, A. Manten, M. van Schothorst, and C. E. Voogd. 1979. Decrease of drug resistance in *Salmonella* in the Netherlands. *Antimicrob. Agents Chemother.* 16:237–239.
42. Varma, J. K., K. Molbak, T. J. Barrett, J. L. Beebe, T. F. Jones, T. Rabatsky-Ehr, K. E. Smith, D. J. Vugia, H. G. Chang, and F. J. Angulo. 2005. Antimicrobial-resistant nontyphoidal *Salmonella* is associated with excess bloodstream infections and hospitalizations. *J. Infect. Dis.* 191:554–561.
43. White, D. G., S. Zhao, R. Sudler, S. Ayers, S. Friedman, S. Chen, P. F. McDermott, S. McDermott, D. D. Wagner, and J. Meng. 2001. The isolation of antibiotic-resistant *Salmonella* from retail ground meats. *N. Engl. J. Med.* 345:1147–1154.
44. Witte, W. 2000. Selective pressure by antibiotic use in livestock. *Int. J. Antimicrob. Agents* 16(Suppl. 1):S19–S24.
45. World Health Organization. 1990. Report of WHO consultation on salmonellosis control in agriculture. World Health Organization, Geneva.
46. Zhao, S., P. F. McDermott, S. Friedman, J. Abbott, S. Ayers, A. Glenn, E. Hall-Robinson, S. K. Hubert, H. Harbottle, R. D. Walker, T. M. Chiller, and D. G. White. 2006. Antimicrobial resistance and genetic relatedness among *Salmonella* from retail foods of animal origin: NARMS retail meat surveillance. *Foodborne Pathog. Dis.* 3:106–117.
47. Zhao, S., P. F. McDermott, S. Friedman, S. Qaiyumi, J. Abbott, C. Kiessling, S. Ayers, R. Singh, S. Hubert, J. Sofos, and D. G. White. 2006. Characterization of antimicrobial-resistant *Salmonella* isolated from imported foods. *J. Food Prot.* 69:500–507.