Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

HHS Public Access

Author manuscript

Microb Drug Resist. Author manuscript; available in PMC 2018 March 01.

Published in final edited form as:

Microb Drug Resist. 2017 March ; 23(2): 188–193. doi:10.1089/mdr.2016.0080.

Characterization of Resistance Genes and Plasmids from Outbreaks and Illness Clusters Caused by *Salmonella* Resistant to Ceftriaxone in the United States, 2011–2012

Jason P. Folster[1,*], **Julian E. Grass**[2], **Amelia Bicknese**[3], **Julia Taylor**[3], **Cindy R. Friedman**[2], and **Jean M. Whichard**[1]

[1]Enteric Diseases Laboratory Branch, Centers for Disease Control and Prevention, Atlanta, GA

[2]Enteric Diseases Epidemiology Branch, Centers for Disease Control and Prevention, Atlanta, GA

[3]IHRC, Inc. Atlanta, GA

Abstract

Salmonella is an important cause of foodborne illness; however, quickly identifying the source of these infections can be difficult, and source identification is a crucial step in preventing additional illnesses. Although most infections are self-limited, invasive salmonellosis may require antimicrobial treatment. Ceftriaxone, an extended-spectrum cephalosporin, is commonly used for treatment of salmonellosis. Previous studies have identified a correlation between the food animal/retail meat source of ceftriaxone-resistant *Salmonella* and the type of resistance gene and plasmid it carries. In this study, we examined seven outbreaks of ceftriaxone-resistant *Salmonella* infections, caused by serotypes Typhimurium, Newport, Heidelberg, and Infantis. All isolates were positive for a plasmid-encoded bla_{CMY} gene. Plasmid incompatibility typing identified five IncI1 and two IncA/C plasmids. Both outbreaks containing bla_{CMY}-IncA/C plasmids were linked to consumption of cattle products. Three of five outbreaks with bla_{CMY}-IncI1 (ST12) plasmids were linked to a poultry source. The remaining IncI1 outbreaks were associated with ground beef (ST20) and tomatoes (ST12). Additionally, we examined isolates from five unsolved clusters of ceftriaxone-resistant *Salmonella* infections and used our plasmid encoded gene findings to predict the source. Overall, we identified a likely association between the source of ceftriaxone-resistant *Salmonella* outbreaks and the type of resistance gene/plasmid it carries.

Introduction

Salmonella is estimated to be the leading cause of bacterial foodborne illness and outbreaks in the United States (1, 2). Infections are commonly due to the consumption of contaminated food (3). However, identifying the source of these infections can be difficult. This is especially important during outbreak investigations where rapid source identification is crucial for preventing additional illnesses. Although most infections are self-limited, invasive salmonellosis may require antimicrobial treatment (3). Ceftriaxone, an extended-

[*]Corresponding author: Jason P. Folster, PhD, National Antimicrobial Resistance Monitoring System, Centers for Disease Control and Prevention, OID/NCEZID/DFWED/EDLB, 1600 Clifton Road, Atlanta GA 30329, gux8@cdc.gov, Ph: (404) 639-4948, Fax: (404) 639-4290.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

spectrum cephalosporin, is commonly used for treatment of salmonellosis in both adults and children, and considerable research has been performed to identify the mechanisms of cephalosporin resistance in *Salmonella* (4).

In the U.S., cephalosporin resistance in *Salmonella* is mainly mediated by AmpC β-lactamases, encoded by *bla*$_{CMY}$ genes (5). These genes are commonly carried on various types of plasmids, which can be characterized by their incompatibility type. Previous work identified that CMY β-lactamase-encoding plasmids in *Salmonella* isolated from humans in the U.S. tended to be large multidrug-resistant (MDR) IncA/C plasmids or single resistance determinant IncI1 plasmids (6). More importantly, *bla*$_{CMY}$-IncI1 plasmids were much more common among *Salmonella* serotypes commonly associated with poultry, for example serotype Heidelberg, while *bla*$_{CMY}$-IncA/C plasmids were more likely to be identified among cattle/beef associated serotypes, for example serotype Newport (7). A follow up study examined *bla*$_{CMY}$ plasmid types among *Salmonella* ser. Typhimurium, a serotype found among diverse sources including poultry and cattle (6). This study found a strong correlation between *bla*$_{CMY}$ plasmid type and the source of Typhimurium isolates, suggesting that *bla*$_{CMY}$ plasmid type may be a useful indicator of possible source of infection during outbreaks.

In the U.S., clusters of human infections caused by enteric bacteria are detected through PulseNet, the national molecular subtyping network, coordinated by the U.S. Centers for Disease Control and Prevention (CDC). Local, state, and federal partners identify the common source of infections for these clusters based on epidemiologic and laboratory investigations. CDC's National Antimicrobial Resistance Monitoring System (NARMS) receives clinical isolates from public health laboratories for antimicrobial susceptibility testing. In this study, we examined clinical isolates from seven outbreaks and five clusters of ceftriaxone-resistant *Salmonella* infections in the United States during 2011–2012 and characterized the ceftriaxone resistance genes and their corresponding plasmids.

Methods

Epidemiologic data source

We defined a cluster as human infections caused by enteric bacteria with matching pulsed-field gel electrophoresis (PFGE) patterns which are more than would be expected during a given time period and geographic area. PulseNet assigns a standardized code to isolates that are associated with each cluster. State and local public health officials interview case-patients with standard questionnaires to collect information regarding the source of their infections. CDC's Outbreak Response and Prevention Branch (ORPB) enteric disease cluster management database was used to find additional information for clusters. We defined an outbreak as the occurrence of two or more cases of a similar illness resulting from a common source. Enteric disease outbreaks are notifiable events that should be reported to the National Outbreak Reporting System (NORS), which is a web-based platform used by local, state, and territorial health departments in the United States to report all foodborne and waterborne disease outbreaks and enteric disease outbreaks transmitted by contact with environmental sources, and infected persons or animals to CDC. Outbreak reports submitted to NORS include information on the implicated source of infection.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Isolate collection and testing

During January 2011–June 2011, CDC asked state public health laboratories to submit representative clinical isolates from single-state outbreaks caused by *Salmonella* serotypes Enteritidis, Newport, and Typhimurium to NARMS. Additionally, the ten participating health departments in CDC's Foodborne Diseases Active Surveillance Network (FoodNet) were asked to submit isolates from single-state outbreaks caused by any *Salmonella* serotype (8). Beginning in July 2011, CDC asked all participating public health laboratories to submit isolates from single-state outbreaks of all *Salmonella* serotypes. For multistate outbreaks, CDC contacted states involved to request isolates. The PulseNet-assigned standardized code for each isolate was used to link to epidemiologic data collected in NORS and to resistance data collected in NARMS.

Antimicrobial susceptibility testing was performed by broth microdilution (Sensititre®, Cleveland, Ohio) and used to determine minimum inhibitory concentrations for 15 antimicrobial agents; ampicillin, amoxicillin-clavulanic acid, azithromycin, cefoxitin, ceftiofur, ceftriaxone, chloramphenicol, ciprofloxacin, gentamicin, kanamycin, nalidixic acid, streptomycin, sulfisoxazole, tetracycline, and trimethoprim-sulfamethoxazole (9). Resistance was defined by the Clinical and Laboratory Standards Institute (CLSI) interpretive standards, when available (10). For streptomycin, where no CLSI interpretive criteria for human isolates exist, resistance was defined as ≥64 mg/L. Testing was performed according to the manufacturer's instructions and the following quality control strains; *E. coli* ATCC 25922, *Staphylococcus aureus* ATCC 29213, *Enterococcus faecalis* ATCC 29212, and *Pseudomonas aeruginosa* ATCC 27853.

Two enzyme (*Xba*I and *Bln*I) PFGE was performed according to the CDC PulseNet protocol.(11) Isolates were grown overnight on Trypticase Soy Agar with 5% defibrinated sheep blood (TSA-SB) (Becton Dickinson Biosciences). Bacterial cell concentration was adjusted by diluting with sterile cell suspension buffer (100 mM Tris, 100 mM EDTA, pH 8.0) to a turbidity measurement of 0.48–0.52 (Dade Microscan Tubidity Meter). Agarose-embedded cells were lysed by proteinase K treatment and extensively washed. Agarose plugs containing genomic DNA were digested with 50U of *Xba*I and *Bln*I restriction enzymes (New England Biolabs, Ipswich, MA) and incubated at 37°C for 2 hours. The fragments were then separated by PFGE using a CHEF Mapper (Bio-Rad Laboratories) with the following conditions and reagents: 1% SeaKem Gold agarose in 0.5% TBE buffer, voltage at 6 V/cm, run time at 18 hours with switch times ranging from 2.16 to 63.8 seconds, temperature at 14°C. *Salmonella enterica* ser. Braenderup H9812 was used as a molecular reference marker. PFGE profiles generated were submitted to the PulseNet national database administered by CDC. Gel images were captured using the GelDoc XR system (Bio-Rad Laboratories) and Quantity One 1-D analysis software (Bio-Rad Laboratories). Pattern analysis and UPGMA dendrogram generation were performed using BioNumerics software (Applied Maths, Saint-Martens-Latem, Belgium) with the Dice coefficient and tolerance of 1.5%.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Isolate selection

We selected one representative isolate from each cluster or outbreak of ceftriaxone-resistant *Salmonella* from 2011 and 2012, based on the PulseNet-assigned standardized code. After these isolates were further characterized, we queried NORS to determine the source of the outbreak associated with each and the enteric disease cluster management database to determine suspected sources.

PCR amplification of bla_{CMY}

For each isolate, DNA template for PCR was prepared by lysing the bacteria at 95°C and collecting the supernatant following centrifugation for 10 min at 20,000 g (Sorvall RC5B Plus, SS-34 rotor, Thermo Fischer Scientific Inc., Waltham, MA). PCR reactions contained 2x HotStar PCR Master Mix (Qiagen Inc., Valencia, CA), 0.4μM of each primer, 5μl template DNA and sterile PCR water to a final volume of 50μl. Thermal cycling was performed using the following conditions: 15 min at 95˚C, followed by 30 cycles of 95˚C for 30 s, 56˚C for 30 s and 72˚C for 90 s. To determine the presence of bla_{CMY} genes, primers *ampC*1 (5′-ATGATGAAAAAATCGTTATGC-3′) and *ampC*2 (5′-TTGCAGCTTTTCAAGAATGCGC-3′) were used (5). PCR products were electrophoresed and visualized in 1.5% agarose gels containing 2.5% GelRed (Biotium Inc., Hayward, CA). Gel images were captured using the GelDoc XR system (Bio-Rad Laboratories) and Quantity one 1-D analysis software (Bio-Rad Laboratories).

Plasmid purification and characterization

Purified plasmid DNA was used to transform laboratory *Escherichia coli*, in order to separate the bla_{CMY} plasmids from other plasmid types prior to replicon typing, and for plasmid multi-locus sequence typing (pMLST). Plasmids were purified using the QiaFilter Midi kit (Qiagen Inc.), following a modified manufacturer's protocol (12). Electroporation of each plasmid into *E. coli* DH10B Electromax competent cells (Invitrogen, Carlsbad, CA) was performed as previously described (7). Cells were plated on LB agar plates containing 100 mg/L of ampicillin or 4 mg/L ceftriaxone (Sigma-Aldrich, St. Louis, MO). Plasmids were re-purified from a single bla_{CMY} PCR-positive transformant to isolate a single plasmid from each isolate. Purification was performed as described above with the additional modification of growing the cells overnight in 25 ml of LB broth with 100 mg/L of ampicillin or 4 mg/L ceftriaxone. Antimicrobial susceptibility of transformants was performed as described above. Plasmid PCR-based replicon typing (PBRT) was performed as previously described (13). Plasmid multi-locus sequence typing was performed on IncI1 plasmids as previously described (14). Sequencing was performed using Big Dye version 3.1 (Applied Biosystems, Foster City, CA) and sequence reactions were cleaned with Centri-sep plates (Princeton Separations, Adelphia, NJ). The reactions were electrophoresed through POP-7 polymer (Applied Biosystems) on a 3730 DNA Analyzer (Applied Biosystems) equipped with a 48-capillary, 50 cm array. Sequence analysis was performed using Lasergene 8 software (DNASTAR Inc, Madison, WI). Sequences were submitted to the plasmid multi locus sequence type (pMLST) web page (http://pubmlst.org/plasmid/) and the sequence type (ST) was determined.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Results

From 2011–2012, we tested isolates from 114 *Salmonella* clusters or outbreaks, 12 (10.5%) of which were resistant to ceftriaxone. Among these 12, seven met the definition of an outbreak (Table 1) and five were clusters with an unknown source (Table 2).

Of the 7 ceftriaxone-resistant *Salmonella* outbreaks, four serotypes were identified, including Heidelberg, Infantis, Newport, and Typhimurium. All seven isolates were PCR-positive for a *bla*$_{CMY}$ gene, which confers resistance to ceftriaxone (and additional β-lactams) and all seven *bla*$_{CMY}$ genes were plasmid-encoded (Table 1). Plasmid incompatibility typing of purified *bla*$_{CMY}$-plasmids identified five IncI1 plasmids and two IncA/C plasmids. Antimicrobial susceptibility testing of the transformants showed that all of the *bla*$_{CMY}$-IncA/C plasmids conferred an MDR phenotype, with additional resistance to chloramphenicol, sulfisoxazole, and tetracycline (and possibly streptomycin). In all seven isolates, phenotypic resistance was conferred by the *bla*$_{CMY}$-encoding plasmids. Plasmid type was not restricted to serotype, as serotype Typhimurium had representatives that contained IncA/C or IncI1 plasmid types. Among five IncI1 positive outbreaks, consumption of poultry products or contact with live poultry was implicated in three (60%); all three IncI1 plasmids were ST12. The remaining two IncI1 outbreaks were associated with beef (ST20) and tomatoes (ST12). Of the two *bla*$_{CMY}$-IncA/C positive outbreaks, both implicated cattle products (beef). All of the *bla*$_{CMY}$-IncI1 plasmids conferred only the *bla*$_{CMY}$-associated resistance phenotype, suggesting it was the only antimicrobial resistance determinant located on the plasmids.

Among the five unsolved clusters of ceftriaxone-resistant *Salmonella* infections (Table 2), four serotypes were identified, including Enteritidis, Newport, Thompson, and Typhimurium. All five representative isolates from these clusters were PCR-positive for a *bla*$_{CMY}$ gene and all were plasmid-encoded. Plasmid incompatibility typing of purified *bla*$_{CMY}$-plasmids identified three IncA/C plasmids, one IncI1 plasmid, and one IncK/B plasmid. The *bla*$_{CMY}$-IncI plasmid (ST12) and the *bla*$_{CMY}$-IncK/B conferred only the *bla*$_{CMY}$-associated resistance phenotype, suggesting it was the only antimicrobial resistance determinant located on the plasmids. Antimicrobial susceptibility testing of the transformants showed that all *bla*$_{CMY}$-IncA/C plasmids conferred an MDR phenotype. All five isolates all phenotypic resistance observed was conferred by the *bla*$_{CMY}$-encoded plasmids.

Discussion

The identification and characterization of resistance determinants and the plasmids which carry them may be an additional tool in identifying the source of *Salmonella* outbreaks and particularly helpful in unsolved clusters. For the seven solved ceftriaxone-resistant *Salmonella* outbreaks, we found a correlation of resistance determinants and plasmids with the source of the outbreaks (i.e. *bla*$_{CMY}$-IncA/C with beef and *bla*$_{CMY}$-IncI1 with poultry). For the five clusters, we were able to predict a source of infection, based on the resistance determinants and plasmids that were identified, but this is impossible to confirm without additional epidemiological data.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Of the two ceftriaxone-resistant *Salmonella* outbreaks containing *bla*$_{CMY}$-IncA/C plasmids, both were linked to consumption of cattle products (beef). Two clusters of an unknown source were serotype Newport, and had the plasmid type (IncA/C) and the resistance phenotype (AmpC-ACSSuT) that are indicative of an MDR-AmpC Newport strain, which has been common among cattle and outbreaks associated with consumption of beef products (15, 16). Another cluster of an unknown source was serotype Typhimurium, a serotype associated with multiple food and environmental sources, but this isolate contained the same *bla*$_{CMY}$-IncA/C plasmid and AmpC-ACSSuT phenotype, which has commonly been linked to consumption of beef or exposure to cattle (6).

Three of the outbreaks linked to poultry sources had isolates with *bla*$_{CMY}$-IncI1 (ST12) plasmids, which are common to poultry; as did one cluster of an unknown source (17). Moreover, this cluster was caused by serotype Thompson, a serotype that is commonly found among poultry. Given this plasmid/sequence type and serotype combination, it suggests a poultry vehicle (18). The outbreak due to serotype Newport that was linked to tomatoes also contained a *bla*$_{CMY}$-IncI1 (ST12) plasmid. While serotype Newport is commonly associated with cattle, this plasmid type and resistance pattern is not typical of Newport from cattle (MDR-AmpC). Serotype Newport has also been found in previous outbreaks linked to tomatoes (19). These Newport outbreaks due to tomatoes may be caused by on farm contamination, with diverse sources of environmental *Salmonella*, including wild birds (19). Most of the previous *Salmonella* Newport outbreak isolates linked to tomatoes have been susceptible to cephalosporins; however the Newport from this outbreak was drug-resistant (19). Another outbreak, caused by serotype Typhimurium from ground beef, contained isolates which had a *bla*$_{CMY}$-IncI1 plasmid. This does not correlate with previous studies which show that this plasmid type is usually associated with poultry (6). However, the *bla*$_{CMY}$-IncI1 plasmid was ST20, which is not a common ST for IncI1 plasmids from poultry. In fact, we have only seen this ST once previously and it was a *bla*$_{CMY}$-IncI1 plasmid from a clinical isolate of *E. coli* O157:H7 (20). The epidemiologic investigation of this outbreak identified the food vehicle as ground beef from a specific grocery store. However, the store in question was grinding a variety of meat (not only beef) on the premises and grind/cleaning records were incomplete. It is possible that the *Salmonella* serotype Typhimurium may have come from cross-contamination from another source or may have acquired the *bla*$_{CMY}$-IncI1 plasmid from another Enterobacteriaceae in the meat, possibly an *E. coli*. Lastly, one cluster of unknown source was serotype Enteritidis and contained a *bla*$_{CMY}$-IncK/B plasmid. This was the first time we have observed a *bla*$_{CMY}$-IncK/B plasmid among U.S. *Salmonella*, making it impossible for us to predict a source. However, *bla*$_{CMY}$-IncK/B plasmids have been observed among *E. coli* in Canada from both clinical and environmental sources (beaches and drinking water), suggesting that it may not be linked to a specific source (21, 22).

Overall, we identified concordance between the source of infection in ceftriaxone-resistant *Salmonella* outbreaks and the type of resistance gene/plasmid. IncA/C plasmids tend to indicate a cattle source and IncI1 plasmids tend to indicate a poultry source, but sequence types of IncI1 other than ST12 may indicate sources other than poultry. While this supports our hypothesis from our previous studies, we were limited by only being able to examine a small number of outbreaks. We used resistance determinants and plasmids to predict the

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

source of the five unsolved clusters. It should also be noted that while this investigation utilized more traditional molecular techniques (e.g. antimicrobial susceptibility testing, PCR, plasmid purification and plasmid incompatibility typing), these methods tend to be more labor intensive than the newer next generation sequencing technologies and whole genome sequencing (WGS) analysis, which could have generated data quicker. Both CDC-PulseNet and some local and state health departments are beginning to perform WGS of *Salmonella* in real-time which will allow for more rapid molecular investigation of outbreaks. This will help public health and regulatory agencies to quickly identify the sources of antimicrobial resistant outbreaks so that rapid action can be taken to prevent additional infections. While a specific antimicrobial resistance gene and plasmid type may be useful to help identify food sources in future antimicrobial resistant *Salmonella* outbreaks, it is only one aspect of an outbreak investigation. For each outbreak, many facets need to be considered including patient exposure and other epidemiologic data, serotype, PFGE analysis, antimicrobial resistance data, and resistance gene/plasmid type.

Acknowledgments

We thank the NARMS participating public health laboratories for submitting the isolates and PulseNet for identifying outbreaks. We also thank Matt Wise and Kelly Walsh for their assistance in providing the outbreak data. This work was partially supported by an interagency agreement between CDC, USDA, and the FDA Center for Veterinary Medicine.

The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the CDC.

References

1. Scallan E, Hoekstra RM, Angulo FJ, Tauxe RV, Widdowson M-A, Roy SL, Jones JL, Griffin PM. Foodborne illness acquired in the United States--major pathogens. Emerging infectious diseases. 2011; 17:7–15. [PubMed: 21192848]
2. Prevention CfDCa. Surveillance for foodborne disease outbreaks, United States, 2013. Annual Report. 2015
3. Hohmann EL. Nontyphoidal salmonellosis. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America. 2001; 32:263–269. [PubMed: 11170916]
4. Larry, KP., editor. RedBook. Salmonella Infections. Red Book. American Academy of Pediatrics; 2012. p. 635-640.
5. Winokur PL, Vonstein DL, Hoffman LJ, Uhlenhopp EK, Doern GV. Evidence for transfer of CMY-2 AmpC beta-lactamase plasmids between Escherichia coli and Salmonella isolates from food animals and humans. Antimicrob Agents Chemother. 2001; 45:2716–2722. [PubMed: 11557460]
6. Folster JP, Tolar B, Pecic G, Sheehan D, Rickert R, Hise K, Zhao S, Fedorka-Cray PJ, McDermott P, Whichard JM. Characterization of blaCMY plasmids and their possible role in source attribution of Salmonella enterica serotype Typhimurium infections. Foodborne Pathog Dis. 2014; 11:301–306. [PubMed: 24484290]
7. Folster JP, Pecic G, McCullough A, Rickert R, Whichard JM. Characterization of bla(CMY)-Encoding Plasmids Among Salmonella Isolated in the United States in 2007. Foodborne Pathog Dis. 2011; doi: 10.1089/fpd.2011.0944
8. Prevention CfDCa. Foodborne Diseases Active Surveillance Network (FoodNet): FoodNet Surveillance Report for 2013 (Final Report). Atlanta, Georgia: 2015.
9. Prevention CfDCa. National Antimicrobial Resistance Monitoring System for Enteric Bacteria (NARMS): Enteric Bacteria Annual Report. 2013.
10. CLSI. CLSI Document M100-S25. Clinical and Laboratory Standards Institute; 2015. Performance Standards for Antimicrobial Susceptibility Testing; Twenty-first Informational Supplement.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

11. Ribot EM, Fair MA, Gautom R, Cameron DN, Hunter SB, Swaminathan B, Barrett TJ. Standardization of pulsed-field gel electrophoresis protocols for the subtyping of Escherichia coli O157:H7, Salmonella, and Shigella for PulseNet. Foodborne Pathog Dis. 2006; 3:59–67. [PubMed: 16602980]
12. Folster JP, Pecic G, Bolcen S, Theobald L, Hise K, Carattoli A, Zhao S, McDermott PF, Whichard JM. Characterization of extended-spectrum cephalosporin-resistant Salmonella enterica serovar Heidelberg isolated from humans in the United States. Foodborne Pathog Dis. 2010; 7:181–187. [PubMed: 19785533]
13. Carattoli A, Bertini A, Villa L, Falbo V, Hopkins KL, Threlfall EJ. Identification of plasmids by PCR-based replicon typing. J Microbiol Methods. 2005; 63:219–228. [PubMed: 15935499]
14. Garcia-Fernandez A, Chiaretto G, Bertini A, Villa L, Fortini D, Ricci A, Carattoli A. Multilocus sequence typing of IncI1 plasmids carrying extended-spectrum beta-lactamases in Escherichia coli and Salmonella of human and animal origin. J Antimicrob Chemother. 2008; 61:1229–1233. [PubMed: 18367460]
15. Greene SK, Stuart AM, Medalla FM, Whichard JM, Hoekstra RM, Chiller TM. Distribution of multidrug-resistant human isolates of MDR-ACSSuT Salmonella Typhimurium and MDR-AmpC Salmonella Newport in the United States, 2003–2005. Foodborne Pathog Dis. 2008; 5:669–680. [PubMed: 18851677]
16. Prevention CfDCa. Outbreak of multidrug-resistant Salmonella enterica serotype Newport infections associated with consumption of unpasteurized Mexican-style aged cheese--Illinois, March 2006-April 2007. MMWR Morb Mortal Wkly Rep. 2008; 57:432–435. [PubMed: 18437118]
17. Folster JP, Pecic G, Singh A, Duval B, Rickert R, Ayers S, Abbott J, McGlinchey B, Bauer-Turpin J, Haro J, Hise K, Zhao S, Fedorka-Cray PJ, Whichard J, McDermott PF. Characterization of extended-spectrum cephalosporin-resistant Salmonella enterica serovar Heidelberg isolated from food animals, retail meat, and humans in the United States 2009. Foodborne Pathog Dis. 2012; 9:638–645. [PubMed: 22755514]
18. Roy P, Dhillon AS, Lauerman LH, Schaberg DM, Bandli D, Johnson S. Results of salmonella isolation from poultry products, poultry, poultry environment, and other characteristics. Avian Dis. 2002; 46:17–24. [PubMed: 11922330]
19. Greene SK, Daly ER, Talbot EA, Demma LJ, Holzbauer S, Patel NJ, Hill TA, Walderhaug MO, Hoekstra RM, Lynch MF, Painter JA. Recurrent multistate outbreak of Salmonella Newport associated with tomatoes from contaminated fields, 2005. Epidemiol Infect. 2008; 136:157–165. [PubMed: 17475091]
20. Folster JP, Pecic G, Stroika S, Rickert R, Whichard JM. Changing plasmid types responsible for extended-spectrum cephalosporin resistance in Escherichia coli O157:H7 in the USA, 1996–2009. Journal of Global Antimicrobial Resistance. 2014; 2:87–91.
21. Baudry PJ, Mataseje L, Zhanel GG, Hoban DJ, Mulvey MR. Characterization of plasmids encoding CMY-2 AmpC beta-lactamases from Escherichia coli in Canadian intensive care units. Diagn Microbiol Infect Dis. 2009; 65:379–383. [PubMed: 19775846]
22. Mataseje LF, Neumann N, Crago B, Baudry P, Zhanel GG, Louie M, Mulvey MR. Group AROWS. Characterization of cefoxitin-resistant Escherichia coli isolates from recreational beaches and private drinking water in Canada between 2004 and 2006. Antimicrob Agents Chemother. 2009; 53:3126–3130. [PubMed: 19398647]

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Table 1

Characterization of the *bla*$_{CMY}$ plasmids identified among *Salmonella* outbreak isolates

Representative isolate	State/year	Serotype	Source of infection[a]	Plasmid type	Antimicrobial resistance[b]	Sequence type
2011K-1273	CA/2011	Typhimurium	Beef carne asada	IncA/C	AMC AMP AXO FOX TIO CHL FIS STR TET	-
2012K-1456	CA/2012	Typhimurium	Beef stew	IncA/C	AMC AMP AXO FOX TIO CHL FIS STR TET	-
2012K-1420	WA/2012	Heidelberg	Chicken	IncI1	AMC AMP AXO FOX TIO	ST12
2011K-1230	NY/2011	Heidelberg	Chicken livers	IncI1	AMC AMP AXO FOX TIO	ST12
2012K-0593	SD/2012	Infantis	Live Poultry	IncI1	AMC AMP AXO FOX TIO	ST12
2011K-1828	VT/2011	Typhimurium	Ground Beef	IncI1	AMC AMP AXO FOX TIO	ST20
2012K-0781	PA/2012	Newport	Tomatoes	IncI1	AMC AMP AXO FOX TIO	ST12

[a]Source of infection reported to NORS

[b]Resistance conferred by the plasmids. AMC, amoxicillin/clavulanic acid; AMP, ampicillin; AXO, ceftriaxone; FOX, cefoxitin; TIO, ceftiofur; CHL, chloramphenicol; FIS, sulfisoxazole; KAN, kanamycin; STR, streptomycin; TET, tetracycline. Additionally, all transformants were resistant to streptomycin due to the natural resistance of DH10B cells.

Microb Drug Resist. Author manuscript; available in PMC 2018 March 01.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Table 2

Characterization of *bla*$_{CMY}$ plasmids identified among isolates from clusters of *Salmonella* infections

Representative isolate	State/year	Serotype	Source of infection[a]	Plasmid type	Antimicrobial resistance[b]	Sequence type
2012K-0664	CA/2012	Newport	Unknown	IncA/C	AMC AMP AXO FOX TIO CHL FIS STR TET	-
2012K-0135	UT/2012	Newport	Unknown	IncA/C	AMC AMP AXO FOX TIO CHL FIS STR TET	-
2012K-0505	CA/2012	Typhimurium	Unknown	IncA/C	AMC AMP AXO FOX TIO CHL FIS STR TET	-
2012K-0725	IA/2012	Thompson	Unknown	IncI1	AMC AMP AXO FOX TIO	ST12
2012K-0283	MA/2012	Enteritidis	Unknown	IncK/B	AMC AMP AXO FOX TIO	-

[a] Epidemiologic investigations were unable to implicate a source of infection

[b] Resistance conferred by the plasmids. AMC, amoxicillin/clavulanic acid; AMP, ampicillin; AXO, ceftriaxone; FOX, cefoxitin; TIO, ceftiofur; CHL, chloramphenicol; FIS, sulfisoxazole; KAN, kanamycin; STR, streptomycin; TET, tetracycline. Additionally, all transformants were resistant to streptomycin due to the natural resistance of DH10B cells.

Microb Drug Resist. Author manuscript; available in PMC 2018 March 01.