

# Salmonella Enteritidis in Meat, Poultry, and Pasteurized Egg Products Regulated by the U.S. Food Safety and Inspection Service, 1998 through 2003

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## ABSTRACT

The U.S. Food Safety and Inspection Service (FSIS) tests for *Salmonella* in meat, poultry, and egg products through three regulatory testing programs: the Pathogen Reduction–Hazard Analysis and Critical Control Point (PR-HACCP) program, the ready-to-eat program for meat and poultry products, and the pasteurized egg products program. From 1998 through 2003, 293,938 samples collected for these testing programs were analyzed for the presence of *Salmonella enterica* serotypes. Of these samples, 12,699 (4.3%) were positive for *Salmonella*, and 167 (1.3%) of the positive samples (0.06% of all samples) contained *Salmonella* Enteritidis. The highest incidence of *Salmonella* Enteritidis was observed in ground chicken PR-HACCP samples (8 of 1,722 samples, 0.46%), and the lowest was found in steer-heifer PR-HACCP samples (0 of 12,835 samples). *Salmonella* Enteritidis isolates were characterized by phage type, pulsed-field gel electrophoretic pattern, and antimicrobial susceptibility. Phage typing of 94 *Salmonella* Enteritidis isolates identified PT13 (39 isolates) and PT8 (36 isolates) as the most common types. One isolate from a ready-to-eat ham product was characterized as PT4. Electrophoretic analysis of 148 *Salmonella* Enteritidis isolates indicated genetic diversity among the isolates, with 28 unique *Xba*I electrophoretic patterns identified. Of these 148 isolates, 136 (92%) were susceptible to each of 16 antimicrobials tested. Two isolates were resistant to ampicillin alone, and 10 isolates were resistant to two or more antimicrobials. Isolation of *Salmonella* Enteritidis from FSIS-regulated products emphasizes the need for continued consumer education on proper food handling and cooking practices and continued work to decrease the prevalence of *Salmonella* in meat, poultry, and pasteurized egg products.

Infections with nontyphoidal *Salmonella* serotypes cause an estimated 1.4 million illnesses, 15,000 hospitalizations, and over 400 deaths annually (23, 45), with an estimated cost of \$2 billion each year in the United States (15). Mead et al. (23) estimated that 95% of these infections are foodborne. U.S. surveillance data indicate that *Salmonella enterica* serotype Enteritidis accounts for about 20% of all foodborne salmonellosis cases (3). Surveillance data from the Centers for Disease Control and Prevention (CDC) (8) identified *Salmonella* Enteritidis as the second most frequently isolated *Salmonella* serotype causing human infection in both 2002 and 2003. In 2002, 15.8% of *Salmonella* isolates (5,116 of 32,308) from human sources that were reported to the CDC were *Salmonella* Enteritidis, whereas in 2003, 14.5% (4,863 of 33,589) were *Salmonella* Enteritidis (8). From 1990 through 2001, state and territorial health departments reported 677 *Salmonella* Enteritidis outbreaks, accounting for 23,366 illnesses, 1,988 hospitalizations, and 33 deaths (3).

*Salmonella* Enteritidis has been recovered from eggs and from clinically affected and healthy chickens, turkeys, cattle, swine, and other domestic and wild animals (8). Although some known-source outbreaks of *Salmonella* Enteritidis infection have been associated with a wide variety of foods, including produce, fresh-pressed juices, and raw almonds, most known-source outbreaks have been linked to contaminated shell eggs (5, 25, 32). From 1985 through 1999, in 298 (80%) of 371 known-source outbreaks of *Salmonella* Enteritidis infection reported to the CDC, eggs or egg-containing foods were confirmed as the vehicle of infection (27). From 1998 through 2002, 73% of the outbreaks of *Salmonella* Enteritidis infection (60 of 82) in which a food vehicle was positively identified were egg related; chicken prepared without egg ingredients was identified as the vehicle of transmission in 4 (5%) of these 82 outbreaks (4). In 2001, three large outbreaks of *Salmonella* Enteritidis infection were associated with eggs. The same predominant *Salmonella* Enteritidis strain was isolated from an egg farm during traceback investigations related to one of these outbreaks, which had resulted in 688 illnesses (3).

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In a Foodborne Disease Active Surveillance Network (FoodNet) case-control study conducted in 1996, Kimura et al. (18) suggested that chicken consumed outside the home may be a significant risk factor for *Salmonella* Enteritidis infections. Results of a more recent 2002 to 2003 FoodNet case-control study also revealed an association between *Salmonella* Enteritidis infection and the consumption of chicken prepared outside the home (21).

As part of its responsibility for ensuring the safety, wholesomeness, and accurate labeling of meat, poultry, and pasteurized egg products, the U.S. Department of Agriculture (USDA) Food Safety and Inspection Service (FSIS) tests products for the presence of *Salmonella*. Three FSIS regulatory programs include testing for *Salmonella* in meat, poultry, or egg products: (i) the Pathogen Reduction–Hazard Analysis and Critical Control Point (PR-HACCP) verification program, (ii) the ready-to-eat (RTE) meat and poultry products program, and (iii) the pasteurized egg products program.

For the years 1998 through 2003, the FSIS previously reported the isolation of *Salmonella* from randomly selected PR-HACCP samples (the A sample set) of ground turkey (1,530 of 5,791; 26.4%), ground chicken (387 of 1,722; 22.5%), and broiler chickens (5,251 of 47,090; 11.2%) (40). During the same time period, *Salmonella* was isolated from 4.3% (1,288) of 29,976 market hog carcass, 2.8% (3,839) of 134,788 ground beef, 1.9% (242) of 12,884 cow and bull carcass, and 0.4% (50) of 12,835 steer and heifer carcass A set samples. *Salmonella* also was isolated from 0.2% (64) of 38,706 RTE samples (41) and 0.5% (48) of 10,146 pasteurized egg product samples.

Phenotypic and genotypic characterization of *Salmonella* Enteritidis isolates is important because it provides data for attribution studies. Comparisons between human and animal isolates can help establish the approximate association of various food commodities with human illness. The objective of this article is to describe the presence of *Salmonella* Enteritidis in samples recovered from these three FSIS regulatory testing programs from 1998 through 2003 and to report the phage types, pulsed-field gel electrophoresis (PFGE) patterns, and antimicrobial susceptibility profiles. These regulatory programs are not designed to estimate the overall prevalence of *Salmonella* or specific prevalence of *Salmonella* Enteritidis. However, these data provide insight into the burden of illness associated with meat and poultry products.

## MATERIALS AND METHODS

**Regulatory testing programs.** Isolates identified in this study were collected as part of the FSIS regulatory testing of PR-HACCP A set verification samples, RTE meat and poultry product samples, and pasteurized egg product samples for the years 1998 through 2003. The PR-HACCP A set samples comprise the initial set of required samples or a set that follows a passed set within a slaughter or processing facility (30, 31). Isolates identified through FSIS regulatory testing of follow-up (targeted) PR-HACCP B and C set verification samples and investigative samples were excluded from this study (30, 31). A total of 293,938 samples from the three regulatory testing programs were analyzed as part of this study.

**Sample collection and analysis.** All PR-HACCP verification samples were aseptically collected by FSIS inspection program personnel and analyzed in one of the three FSIS field service laboratories as previously described (30, 31). Effective October 2003, the BAX System PCR Assay for Screening *Salmonella* (DuPont Qualicon, Wilmington, Del.) (38) replaced the automated immunoassay screen system (Assurance EIA *Salmonella*, Bio-Control Systems, Inc., Bellevue, Wash.) for screening raw product sample enrichment cultures.

Before December 2000, RTE meat and poultry samples were collected and shipped by FSIS inspection program personnel to one of the three FSIS field service laboratories following sampling protocols that have been previously described (20). Since December 2000, inspectors have been instructed to submit a minimum of 2 lb (0.9 kg) of intact packaged RTE product for laboratory analysis. When an intact product sample exceeds 2 lb, inspectors request that the establishment “slack fill” or reduce the contents of the package to 2 lb. Intact packages were shipped either refrigerated or frozen to the designated FSIS laboratory, where a 325-g sample prepared with portions from each individual package representing a sampled lot of product was analyzed as described (39). In February 2003, the two previously described enzyme-linked immunosorbent assays for screening RTE products (20) were replaced by the BAX System PCR assay (38).

Pasteurized egg product samples were collected and shipped by FSIS inspection program personnel to one of the three FSIS field service laboratories (37). Liquid products were shipped either refrigerated or frozen, and dried products were shipped at ambient temperature. At the laboratory, a 100-g sample from each sampled lot of egg product was prepared and analyzed according to the FSIS culture method (39) without the use of a screen test. In February 2003, the BAX System PCR Assay (38) was added to the egg products analysis.

***Salmonella* serotyping and phage typing.** One suspect *Salmonella* isolate per sample from the dominant colony type present on the culture plate was confirmed biochemically and serologically (polyvalent H and O group antigens and individual O group antigens A through I) (39). Presumptive-positive *Salmonella* isolates were forwarded to the USDA National Veterinary Services Laboratory (NVSL; Ames, Iowa) for serotyping (13). Beginning in 2001, the NVSL phage typed all *Salmonella* Enteritidis isolates (46).

**PFGE.** Isolates were analyzed by PFGE at the USDA VetNet Laboratory (Athens, Ga.) A 24-h *Salmonella* PFGE procedure was performed as described by PulseNet (7). Bacterial genomic DNA was digested with 10 U of *Xba*I (Roche Molecular Biochemicals, Indianapolis, Ind.). DNA standards were prepared from *S. enterica* serotype Newport AM01144. Digested DNA was separated with the CHEF-DRII PFGE system as per the manufacturer’s instructions (Bio-Rad, Hercules, Calif.). Electrophoresis was done at 6 V for 19 h with a ramped pulse time of 2.16 to 63.8 s in 0.5× Tris-borate-EDTA buffer at 14°C. Cluster analysis was applied with the BioNumerics software program (Applied Maths Scientific Software Development, Sint-Martens-Latem, Belgium) using the Dice coefficient and the unweighted pair-group method (UPGMA).

**Antimicrobial susceptibility testing.** Isolates were also submitted to the USDA Agricultural Research Service for antimicrobial susceptibility testing as part of the animal arm of the National Antimicrobial Resistance Monitoring System—Enteric Bacteria (Athens, GA) (34, 35). Antimicrobial susceptibility was determined using a custom-made 96-well panel and a semiautomated

broth microdilution testing system (Sensititre, TREK Diagnostics, Inc., Westlake, Ohio) as per the manufacturer's instructions. Where possible, full-range MICs were used. *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, and *Enterococcus faecalis* 29212 were used as quality control strains. Results were interpreted according to the CLSI (formerly the NCCLS) guidelines available (24).

**Descriptive statistical analyses.** For each product category of broiler chicken, market hog, cow and bull, and steer and heifer carcasses, ground chicken, ground turkey, ground beef, RTE meat, RTE poultry, and pasteurized egg products, the number of *Salmonella* Enteritidis isolations was summarized relative to the number of total *Salmonella* isolations and to the number of samples analyzed. Phage types, PFGE patterns, and antimicrobial susceptibilities of *Salmonella* Enteritidis isolates were summarized by product category.

## RESULTS

**Regulatory sampling.** Of the 293,938 samples analyzed in this study, 12,699 (4.3%) were positive for *Salmonella*, and 167 (1.3% of 12,699 *Salmonella*-positive samples or 0.06% of all 293,938 samples) were positive for *Salmonella* Enteritidis. Classical serotypes were obtained for 92.2% (11,708) of the 12,699 isolates, and 3.4% (437) of these isolates were characterized as monophasic, nonmotile, untypeable, or nonrecoverable. The remaining 4.4% (554) of the *Salmonella* isolates were not serotyped.

*Salmonella* was most often isolated from ground turkey (26.4%; 1,530 of 5,791 samples), ground chicken (22.5%; 387 of 1,722 samples), and broilers (11.2%; 5,251 of 47,090 samples). The highest percentages of *Salmonella* Enteritidis was observed in ground chicken (0.46%; 8 of 1,722 samples) and broiler chickens (0.26%; 124 of 47,090 samples) (Table 1). *Salmonella* Enteritidis was isolated from 0.07% (7) of 10,146 pasteurized egg products samples and 0.005% (2) of 38,706 RTE product samples.

The largest number of *Salmonella* Enteritidis isolates was recovered from broilers (124 positive samples). As a percentage of total *Salmonella*-positive samples by product for all years, *Salmonella* Enteritidis was identified most often in pasteurized egg products (14.58%; 7 of 48 samples), RTE meat and poultry products (3.13%; 2 of 64 samples), broiler chickens (2.36%; 124 of 5,251 samples), and ground chicken (2.07%; 8 of 387 samples) (Table 1). Although the majority of samples (134,788) were of ground beef, 0.47% of the total number of *Salmonella* isolates found in ground beef (in 18 of 3,839 samples) were identified as *Salmonella* Enteritidis. In the group of *Salmonella*-positive samples, *Salmonella* Enteritidis was found in 0.39% (5) of 1,288 market hog samples, 0.41% (1) of 242 cow and bull samples, and 0.13% (2) of 1,530 ground turkey samples.

**Phage types.** NVSL provided phage-typing results for 94 *Salmonella* Enteritidis isolates (Table 2). Of the 69 broiler isolates phage typed, phage type (PT) 13 (36 isolates, 52%) and PT8 (26 isolates, 38%) were the predominant types. The remaining broiler chicken *Salmonella* Enteritidis isolates were PT2, PT4B, PT13A, and PT28. Figure 1 illustrates the number of isolates recovered from broiler samples by phage type for 2001 through 2003. PT8 and PT13

predominated in ground beef, and PT8 predominated in ground chicken and pasteurized egg products. Recovery of other phage types was sporadic. PT4 was not identified in the broiler population sampled; the single PT4 isolate was found in an RTE ham sample.

**PFGE.** PFGE analysis was performed for 148 (88.62%) of the 167 *Salmonella* Enteritidis isolates (Table 3). From the 148 patterns, 28 unique *Xba*I patterns were identified (Fig. 2). Two VetNet pattern designations, JEGX01.0003 and JEGX01.0002, were identified most often among all isolates.

VetNet pattern JEGX01.0003 isolates were recovered from poultry, ground beef, and market hogs, whereas pattern JEGX01.0002 isolates were primarily recovered from poultry (Table 3). Of 112 broiler isolates analyzed, 43 (38.4%) were pattern JEGX01.0003 and 42 (37.5%) were pattern JEGX01.0002. The number of broiler isolates with pattern JEGX01.0003 varied over time, whereas pattern JEGX01.0002 appears to have increased since 2001. VetNet pattern JEGX01.0003 is indistinguishable from PulseNet pattern JEGX01.0004, and VetNet pattern JEGX01.0002 is indistinguishable from PulseNet pattern JEGX01.0005. PulseNet pattern JEGX01.0004 is the most common *Salmonella* Enteritidis pattern in the PulseNet database, with a frequency of 37%, and PulseNet pattern JEGX01.0005 is the third most common, with a frequency of 12.65% (22). Of 18 *Salmonella* Enteritidis isolates from ground beef, 11 (61.1%) were pattern JEGX01.0003. Of five market hog isolates, three (60%) were pattern JEGX01.0003 (Table 3).

**Antimicrobial susceptibility.** Of the 148 *Salmonella* Enteritidis isolates tested, 136 (91.9%) were pansusceptible. Figure 3 shows the PFGE pattern, product, year, and resistance pattern for the 12 isolates exhibiting resistance. Of those 12 isolates, 10 were recovered from broiler chicken carcass samples, 1 was from a market hog carcass, and 1 was from ground chicken. Two isolates were resistant to ampicillin alone, and 10 isolates were resistant to two or more antimicrobials. Of the 12 resistant isolates, 11 were resistant to ampicillin, 3 were resistant to tetracycline, 4 were resistant to sulfamethoxazole, 3 were resistant to cephalothin, and 3 were resistant to ticarcillin. Four of the 12 isolates were resistant to four or more antimicrobials. The most common multiresistance pattern was to AmpTic ( $n = 3$ ) (Fig. 3). Although JEGX01.0002 and JEGX01.0003 were the most common PFGE patterns, resistance appeared to be spread among a number of other patterns, including JEGX01.0010, JEGX01.0013, JEGX01.0015, and JEGX01.0025 (Fig. 3).

## DISCUSSION

As the regulatory agency responsible for ensuring the safety of meat, poultry, and egg products in the United States, the FSIS collects and analyzes samples from all sizes of federally inspected slaughter and processing establishments across the country. The result is a unique and comprehensive data set. Although FSIS sampling programs were designed to verify compliance with food safety regulations and not to measure prevalence, sampling data can

TABLE 1. Samples positive for Salmonella and specifically Salmonella Enteritidis from FSIS regulatory sampling by year and product class

Sample group	Year:							Total
	1998	1999	2000	2001	2002	2003		
<b>PR-HACCP regulatory program</b>								
<b>Broilers</b>								
<i>n</i> <sup>a</sup>	5,659	6,768	10,057	8,955	9,183	6,468	47,090	
No. <i>Salmonella</i> positive <sup>b</sup>	613	772	914	1,065	1,059	828	5,251	
% (no.) <i>Salmonella</i> Enteritidis <sup>c</sup>	2.28 (14)	1.04 (8)	2.52 (23)	1.60 (17)	3.12 (33)	3.50 (29)	2.36 (124)	
<b>Market hogs</b>								
<i>n</i>	1,390	1,923	5,170	8,090	7,479	5,924	29,976	
No. <i>Salmonella</i> positive	81	189	323	307	237	151	1,288	
% (no.) <i>Salmonella</i> Enteritidis	0	0.53 (1)	0.31 (1)	0.33 (1)	0.42 (1)	0.66 (1)	0.39 (5)	
<b>Cows and bulls</b>								
<i>n</i>	179	1,521	1,995	2,176	4,414	2,599	12,884	
No. <i>Salmonella</i> positive	2	33	43	53	73	38	242	
% (no.) <i>Salmonella</i> Enteritidis	0	0	0	1.89 (1)	0	0	0.41 (1)	
<b>Steers and heifers</b>								
<i>n</i>	214	782	1,092	1,695	4,572	4,480	12,835	
No. <i>Salmonella</i> positive	0	2	4	11	14	19	50	
% (no.) <i>Salmonella</i> Enteritidis	0	0	0	0	0	0	0	
<b>Ground beef</b>								
<i>n</i>	1,296	16,375	32,844	24,243	30,933	29,097	134,788	
No. <i>Salmonella</i> positive	83	710	1,080	686	790	490	3,839	
% (no.) <i>Salmonella</i> Enteritidis	0	0.42 (3)	0.37 (4)	0.44 (3)	0.76 (6)	0.41 (2)	0.47 (18)	
<b>Ground chicken</b>								
<i>n</i>	24	297	414	262	429	296	1,722	
No. <i>Salmonella</i> positive	1	48	57	51	125	105	387	
% (no.) <i>Salmonella</i> Enteritidis	0	2.08 (1)	1.75 (1)	0	4.80 (6)	0	2.07 (8)	
<b>Ground turkey</b>								
<i>n</i>	591	1,050	1,551	520	1,075	1,004	5,791	
No. <i>Salmonella</i> positive	216	332	399	136	192	255	1,530	
% (no.) <i>Salmonella</i> Enteritidis	0	0.30 (1)	0.25 (1)	0	0	0	0.13 (2)	
<b>Other regulatory programs</b>								
<b>RTE meat and poultry</b>								
<i>n</i>	3,466	7,130	8,330	6,539	7,582	5,659	38,706	
No. <i>Salmonella</i> positive	8	19	10	10	13	4	64	
% (no.) <i>Salmonella</i> Enteritidis	0	0	10.00 (1)	0	7.69 (1)	0	3.12 (2)	
<b>Pasteurized egg products</b>								
<i>n</i>	1,812	1,710	1,761	1,656	1,647	1,560	10,146	
No. <i>Salmonella</i> positive	10	14	6	6	7	5	48	
% (no.) <i>Salmonella</i> Enteritidis	10.00 (1)	21.43 (3)	0	33.33 (2)	0	20.00 (1)	14.58 (7)	

<sup>a</sup> Number of samples tested.  
<sup>b</sup> Number of *Salmonella*-positive samples.  
<sup>c</sup> Percentage of samples with *Salmonella* Enteritidis of total *Salmonella*-positive samples (number of samples in which *Salmonella* Enteritidis was identified).

TABLE 2. *Phage types of Salmonella Enteritidis isolates from FSIS regulatory product samples by year*

Product <sup>a</sup>	Year <sup>b</sup>	No. of isolates of each phage type:											Total no. of isolates typed
		PT2	PT4	PT4B	PT8	PT13	PT13A	PT23	PT24	PT28	PT34	PT42	
Broilers	2001	1	— <sup>c</sup>	—	4	11	—	—	—	—	—	—	16
	2002	—	—	—	8	12	2	—	—	3	—	25	
	2003	—	—	1	14	13	—	—	—	—	—	28	
Market hogs	2001	—	—	—	—	—	—	—	1	—	—	1	
	2003	—	—	—	1	—	—	—	—	—	—	1	
Ground beef	2001	—	—	—	1	2	—	—	—	—	—	3	
	2002	—	—	—	2	1	—	—	—	1	1	5	
	2003	—	—	—	—	—	—	1	—	1	—	2	
Ground chicken	2002	1	—	—	3	—	—	—	—	1	—	5	
RTE meat and poultry	2000	—	—	—	—	—	—	—	—	1	—	1	
	2002	—	1	—	—	—	—	—	—	—	—	1	
Pasteurized egg products	1998	—	—	—	—	—	—	—	—	1	—	1	
	1999	—	—	—	2	—	1	—	—	—	—	3	
	2001	1	—	—	—	—	—	—	—	—	—	1	
	2003	—	—	—	1	—	—	—	—	—	—	1	

<sup>a</sup> Product classes with no *Salmonella* Enteritidis or phage type data available are not included.

<sup>b</sup> Within each product class, years with no *Salmonella* Enteritidis or phage type data available are not included. Phage types were not routinely determined for PR-HACCP isolates until 2001.

<sup>c</sup> —, no isolates of this phage type were found.

provide useful information on the occurrence of foodborne pathogens in meat, poultry, and egg products (44).

*Salmonella* Enteritidis was isolated from 0.06% (167) of all 293,938 samples and from 1.32% (167) of 12,699 *Salmonella*-positive samples analyzed in this study. Although *Salmonella* Enteritidis is a known egg-associated foodborne pathogen, the highest percentages of *Salmonella* Enteritidis-positive samples were found in ground chicken (0.46%; 8 of 1,722 samples) and broiler chickens (0.26%; 124 of 47,090 samples). This high incidence warrants further investigation at the farm level to determine whether *Salmonella* Enteritidis is becoming more prevalent in broiler chickens.

Recent FSIS data not included in this study indicate an increased percentage of samples positive for *Salmonella* in A sample sets from broiler (young chicken) establishments, from 12.8% in 2003 to 13.5% in 2004 and 16.3% in 2005 (40). A disconcerting proportion of *Salmonella* serotypes commonly associated with human illness was found among the positive samples, including serotypes Enteritidis, Typhimurium, and Heidelberg. In response to these trends, in February 2006 the FSIS announced a new initiative to reduce *Salmonella* contamination in raw meat and poultry

products (42), which included changes in the way FSIS reports and uses data gathered through the *Salmonella* verification sampling program.

Although *Salmonella* Enteritidis is most frequently isolated from broiler samples, it was also isolated from a variety of other animal products. The pathogen was identified in ground beef (18 isolates), pasteurized egg products (7 isolates), market hog carcasses (5 isolates), ground turkey (2 isolates), RTE meat and poultry products (2 isolates), and cow and bull carcasses (1 isolate). These findings suggest that a variety of animal species can harbor *Salmonella* Enteritidis and result in food contamination.

The low isolation rates for *Salmonella* Enteritidis from pasteurized egg products (0.07%; 7 of 10,146 samples) and RTE products (0.005%; 2 of 38,706 samples) indicate a high degree of control in these products. Although these rates are very low, both pasteurized egg products and RTE meat and poultry products could be consumed without additional cooking, which may pose a potentially greater risk of foodborne illness than the consumption of products that are expected to be cooked by consumers.

The proportion of *Salmonella* Enteritidis-contaminated samples in the total *Salmonella*-positive sample set by prod-

FIGURE 1. *Salmonella* Enteritidis phage types from FSIS regulatory broiler samples, 2001 through 2003.

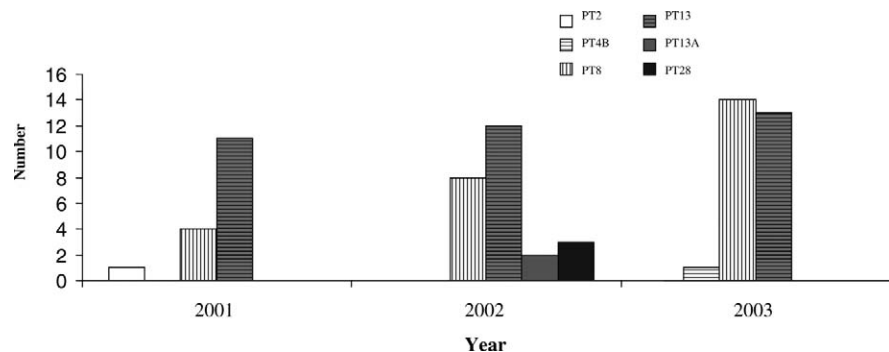


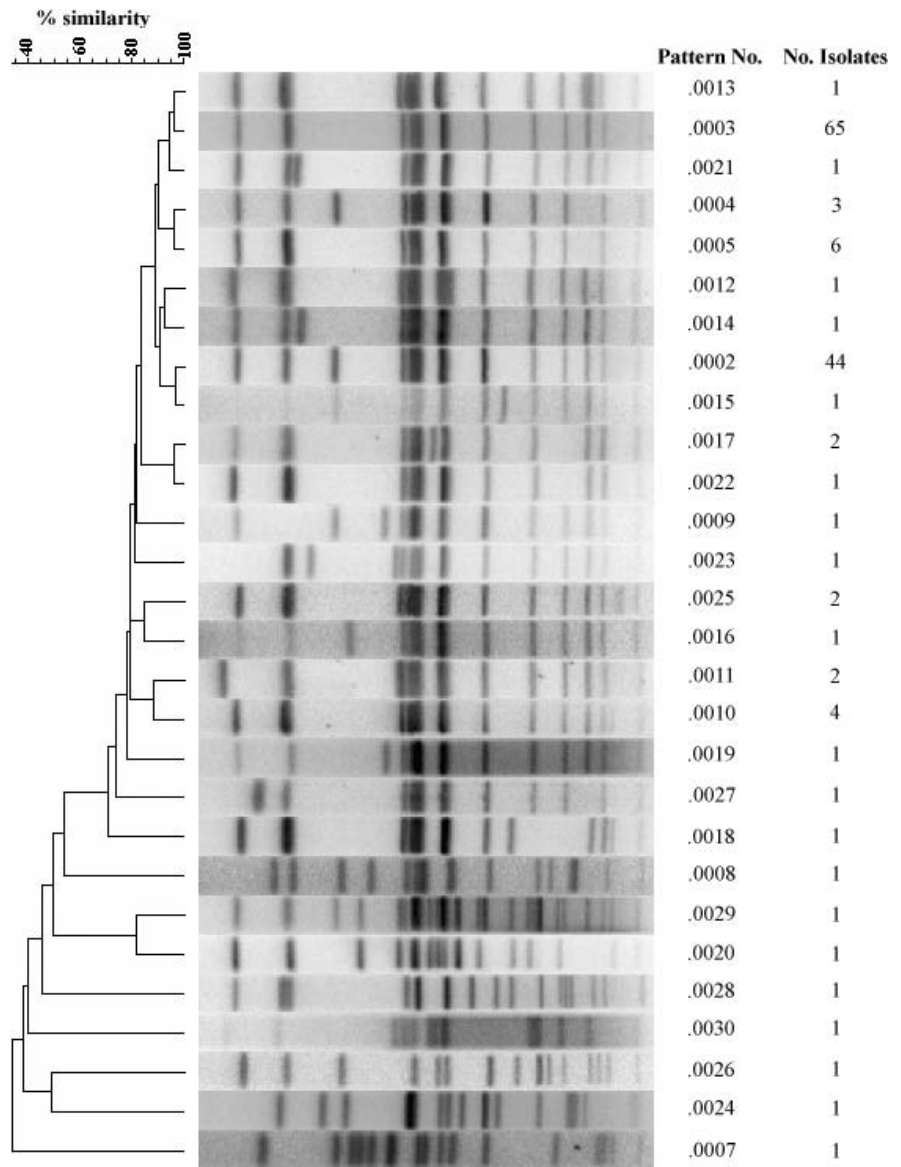
TABLE 3. PFGE patterns from regulatory product samples positive for *Salmonella Enteritidis*

Product	No. of isolates per PFGE pattern <sup>a</sup> :																												
	.0002	.0003	.0004	.0005	.0007	.0008	.0009	.0010	.0011	.0012	.0013	.0014	.0015	.0016	.0017	.0018	.0019	.0020	.0021	.0022	.0023	.0024	.0025	.0026	.0027	.0028	.0029	.0030	
1998																													
Broilers	— <sup>b</sup>	2	—	—	—	—	—	2	—	—	—	—	—	—	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—
1999																													
Broilers	2	3	1	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Market hogs	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground beef	—	1	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground chicken	—	—	—	—	—	—	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground turkey	—	—	—	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
2000																													
Broilers	5	11	—	1	—	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Market hogs	—	—	—	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground beef	—	4	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground chicken	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground turkey	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
2001																													
Broilers	10	4	—	—	—	—	—	—	—	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Market hogs	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Cows and bulls	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground beef	1	2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
2002																													
Broilers	13	14	2	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Market hogs	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground beef	1	2	—	—	—	—	—	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground chicken	—	5	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
RTE meat and poultry	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
2003																													
Broilers	12	9	—	3	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Market hogs	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Ground beef	—	2	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Pasteurized egg product	—	1	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Total	44	65	3	6	1	1	1	1	4	2	1	1	1	1	1	1	1	1	1	1	1	1	1	2	1	1	1	1	1

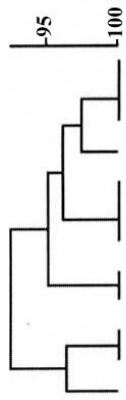
<sup>a</sup> Each PFGE *Xba*I pattern is represented by a 10-character code. The first three characters represent the bacterial pathogen (JEG), the next three characters denote the enzyme used for DNA restriction (X01), and the last four characters represent the VetNet pattern designation. These pattern numbers (shown here) are assigned sequentially to unique patterns by VetNet.

<sup>b</sup> —, no isolates matching the PFGE pattern were found.

FIGURE 2. Most common PFGE patterns among *Salmonella Enteritidis* isolates.



% similarity



Product Class	Year	PFGE Pattern	AR Phenotype
Broiler	1999	.0010	AmpCep
Broiler	1998	.0010	AmpTic
Broiler	1998	.0010	AmpTic
Ground chicken	1999	.0013	AmpTet
Broiler	2000	.0003	Amp
Market hog	2001	.0003	AmpKanStrTet
Broiler	2002	.0003	AmoAmpFoxTioAxoCepSul
Broiler	2000	.0025	Amp
Broiler	1998	.0025	AmpTic
Broiler	2000	.0002	AmoAmpTioSul
Broiler	2003	.0002	SulTet
Broiler	2002	.0015	AmoAmpTicCepKanStrSulTet

FIGURE 3. PFGE patterns from *Salmonella Enteritidis* isolates exhibiting antimicrobial resistance. Amp, ampicillin; Tic, ticarcillin; Cep, cephalothin; Amo, amoxicillin-clavulanic acid; Tio, ceftiofur; Sul, sulfamethoxazole; Fox, ceftioxin; Axo, ceftriaxone; Kan, kanamycin; Str, streptomycin; Tet, tetracycline.

uct for all years was highest in pasteurized egg products (14.6%; 7 of 48 samples). The practice of diverting shell eggs from *Salmonella* Enteritidis–positive layer flocks to egg products increases the likelihood of finding *Salmonella* Enteritidis among the *Salmonella* serotypes isolated from egg products. The FSIS is taking additional steps to ensure that egg products are properly processed and effectively treated to destroy *Salmonella*. The overall FSIS strategy to reduce concentrations of pathogens in processed egg products includes proposing implementation of HACCP and sanitation standard operating procedure regulations and lethality-based performance standards for egg products and pasteurized shell eggs.

Sixty-two of 69 *Salmonella* Enteritidis strains recovered from broiler samples in this study were PT8 and PT13. The *Salmonella* Enteritidis isolates recovered from pasteurized egg products included PT8 (3 of 6 isolates; 50%) and PT13a. Between 1998 and 2002, the CDC Surveillance Summary Data included 214 outbreaks of *Salmonella* Enteritidis infection. Phage typing was performed on patient isolates in 66% (142) of these outbreaks. The dominant phage types isolated during outbreaks included PT4 (27%, 38 isolates), PT8 (25%, 35 isolates), PT13a (18%, 25 isolates), and PT13 (6%, 9 isolates). Twenty-five of the 38 PT4-associated outbreaks occurred within California before 2001 (4, 19). Increases in human infections (2, 26, 32) have been linked to strains of *Salmonella* Enteritidis PT4, which harbor unique virulence characteristics (16, 17, 28). In the United States, *Salmonella* Enteritidis PT4 has been identified in commercial laying flocks (26) and in eggs (2, 26, 32), but unlike in Europe (12, 29), it has not been reported in U.S. broiler chickens. Although in a previous case-control study (18) the researchers postulated that *Salmonella* Enteritidis PT4 may be present in broiler flocks in the United States, the lack of this pathogen type in broiler carcasses in this study should help allay those concerns. The sole PT4 isolate identified in this study was found in an RTE sliced ham product. The presence of *Salmonella* in an RTE product is unusual, as is isolation of *Salmonella* Enteritidis PT4 from a ham product. Because no poultry was processed at the establishment from which this isolate came, the finding may indicate cross-contamination from a human source shedding *Salmonella* Enteritidis while handling products. Contamination of RTE products with *Salmonella* Enteritidis by establishment workers after lethality treatments should be taken into consideration.

PFGE is an important molecular genetic subtyping tool for characterizing foodborne pathogenic bacteria and is used to detect disease clusters, identify common-source outbreaks, and target resources and mitigations to reduce foodborne illness (7). Characterization of 148 *Salmonella* Enteritidis isolates by PFGE resulted in 28 unique PFGE patterns, which suggests diversity among the isolates. However, most of the isolates had one of two VetNet patterns, JEGX01.0002 and JEGX01.0003, indicating that these are the dominant clones within the population. Isolates exhibiting these PFGE patterns were predominantly recovered from broilers, which with the phage typing data suggest the presence of specific *Salmonella* Enteritidis clones among

the broiler samples from this study. Spread of a limited number of clones among broilers may limit the discriminatory power of PFGE for distinguishing among outbreaks of *Salmonella* Enteritidis infection. Success in distinguishing *Salmonella* Enteritidis isolates implicated in foodborne illness may thus require molecular subtyping methods other than PFGE (17). However, comparisons of PFGE patterns of isolates from food products and from human illness can help to attribute *Salmonella* Enteritidis infections to a food category.

All but 12 of the *Salmonella* Enteritidis isolates were susceptible to each of the 16 antimicrobials tested in this study, a finding generally consistent with previous data indicating that a majority of *Salmonella* Enteritidis are pansusceptible (6). None of the isolates were resistant to ciprofloxacin, and only one isolate was resistant to ceftriaxone, drugs frequently used for treating invasive salmonellosis in adults and children, respectively, in the United States (1, 11). The observation elsewhere of emerging phenotypes resistant to third-generation cephalosporins, fluoroquinolones, and other clinically relevant antimicrobials (9, 10, 48) and the fact that cases of antimicrobial-resistant salmonellosis can be acquired by consuming foods of animal origin (1, 14, 35) warrant continued surveillance to detect emerging resistant phenotypes of *Salmonella* Enteritidis from meat, poultry, and eggs. Although there was also a lack of multiple resistance among the isolates, the appearance of multiple resistance to four or more antimicrobials warrants further molecular analysis to determine whether mobile genetic elements (11, 33, 47) are moving into the serotype.

We were unable to make statistical inferences with these data. The microbiological testing programs described in this study are regulatory in nature and designed to track establishment performance and to encourage industry to continually monitor and improve production practices. Thus, these programs were not designed to collect data appropriate for estimating the prevalence of *Salmonella* or specifically *Salmonella* Enteritidis or for monitoring temporal trends in *Salmonella* contamination. For example, during the first 3 years of PR-HACCP implementation (1998 through 2000), the requirements for HACCP systems and *Salmonella* verification testing of the seven raw product categories were phased in based on establishment size. It was not until 2000 that all establishments, regardless of size, were eligible for PR-HACCP testing. Sampling frequency was not proportional to an establishment's production volume and was not stratified by the geographic location of an establishment. However, these data provide information that can guide additional research and identify new opportunities to ensure a safe food supply and protect public health.

Despite the overall low rates of *Salmonella* Enteritidis contamination, high rates of poultry consumption in the United States (36) may contribute to the burden of human illness associated with this pathogen. Recent case-control studies have implicated consumption of chicken outside the home as a risk factor for sporadic cases of *Salmonella* Enteritidis–related illness. In an investigation conducted in 2005 and 2006 of *Salmonella* Enteritidis patients in Min-



nesota, raw stuffed chicken products that appeared fully cooked to the consumer were identified as a source of the infections (43). These findings emphasize how proper food handling practices at the retail level and thorough cooking of raw meat and poultry products play an important role in preventing salmonellosis and other foodborne illnesses. The FSIS is requiring new labels for these stuffed chicken products that clearly state that they contain raw chicken and must be fully cooked to a safe minimum internal temperature of 165°F (73.9°C).

Efforts to reduce human infection from foodborne *Salmonella* Enteritidis and other pathogens must focus on the implementation of farm-to-table prevention strategies. Pre-harvest interventions such as the National Poultry Improvement Plan SE Clean program for meat-type breeders, best production practices (quality assurance programs), and microbiological monitoring in the grow-out facility are essential prevention strategies. Proper implementation and design of PR-HACCP systems and compliance with the *Salmonella* performance standards by federally regulated meat and poultry establishments help to reduce human exposure to salmonellae, including *Salmonella* Enteritidis (44). The initiatives announced by the FSIS in February 2006 will help reduce *Salmonella* in raw meat and poultry products by dedicating additional FSIS resources to establishments that have demonstrated poor process control. The FSIS will modify the scheduling frequency for *Salmonella* sample sets and will conduct food safety assessments in establishments that fail to control *Salmonella* contamination (42).

The FSIS is also changing how it will report these results. Instead of reporting only the results of completed sample sets, the FSIS will report results of each individual *Salmonella* test, thus allowing establishments to assess the results and make adjustments to their food systems accordingly. The FSIS also plans to pursue more timely mechanisms for obtaining serotype information for each *Salmonella* verification sample. Establishments will be given this serotype information, and the FSIS will publish aggregate results at least annually. There will be a particular focus on sample sets that contain serotypes that are common causes of human illness, such as *Salmonella* Enteritidis (42).

The FSIS expects that these actions will encourage additional industry-wide efforts to control *Salmonella*. To ascertain continuous improvements in pathogen reduction in all classes or raw product, the FSIS will conduct on-going baseline studies. These studies will assess changes in serotypes and patterns of antibiotic resistance and will monitor pathogen levels in federally regulated meat and poultry products.

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