

Survey of Not Ready-to-Eat Breaded and Stuffed Chicken Products for *Salmonella*

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Executive Summary

While the prevalence of *Salmonella* contamination in regulated poultry products has decreased by more than 50 percent in recent years, there has not been a reduction in human illnesses attributable to poultry. Over one million *Salmonella* illnesses occur annually, with more than 23 percent of foodborne salmonellosis attributed to poultry consumption. From 1998 to 2021, FSIS and public health partners have investigated 14 *Salmonella* illness outbreaks associated with consumption of not ready-to-eat breaded and stuffed chicken products. To learn more, FSIS conducted a non-scientific study to determine the presence of *Salmonella* in these products.

FSIS worked with the Food Emergency Response Network to test for the presence of *Salmonella* and sanitary indicator aerobic organism counts using the current validated methods employed by 11 state public health and agriculture laboratories. From July 1, 2022, to September 30, 2022, the laboratories purchased approximately 15 samples of the product per month from nearby retail locations.

In total, 58 of the 487 samples were positive for the presence of *Salmonella*. A noticeable difference in positivity rates was seen between laboratories that ran methods the same as FSIS (36 of 135, 27%) using larger test portions and laboratories that ran methods different from FSIS (22 of 352, 6%) using smaller test portions. Small amounts of *Salmonella* present in the product or uneven distribution of *Salmonella* contamination could account for the lower positivity rates when smaller sample portions were tested. Total aerobic counts were similar across products tested. Whole genome sequencing of *Salmonella* detected *Salmonella* Enteritidis (18/58), *Salmonella* Infantis (22/58), *Salmonella* Kentucky (15/58) and *Salmonella* Typhimurium (3/58).

Introduction

FSIS conducted a study with the Food Emergency Response Network (FERN) Cooperative Agreement Laboratories to gather data on NRTE breaded pre-browned stuffed chicken products (“NRTE stuffed chicken”) purchased at retail. Specifically, FSIS was interested in gaining information on the positive rate of *Salmonella* in these products and any differences in testing strategies. Through the FERN, 11 geographically dispersed state laboratories participated in this study. From July 1, 2022, to September 30, 2022, these laboratories purchased locally available NRTE stuffed chicken products at retail and tested for the presence of *Salmonella* and sanitary indicator aerobic organism counts using current validated methods.

Methodology

Samples: FSIS provided the laboratories with an information sheet with visual depictions of the appropriate NRTE stuffed chicken products to purchase. Laboratories were encouraged to find samples from as many different brands as were available to them. All samples were purchased at retail locations surrounding the laboratories. Participating laboratories included the California Department of Public Health Food and Drug Laboratory, Colorado Department of Agriculture, Florida Department of Agriculture, State Hygienic Laboratory of Iowa, Michigan Department of Health and Human Services, Minnesota Department of Agriculture, Missouri Department of Health, New York Department of Health Wadsworth Center, Ohio Department of Agriculture, Texas State Chemist Laboratory, and Virginia Division of Consolidated Laboratory Services. The USDA establishment numbers on each package were recorded, and establishments were referred to by a letter designation (i.e., blinded) when results were reported to FSIS.

Methods: Laboratories used methods listed under their current ISO accreditation scope. Three laboratories followed the FSIS Microbiological Laboratory Guidebook (MLG) method, which uses a sample portion of at least 325g. One of the laboratories utilized a 375g sample: this method has been given equivalent status through the FSIS State Meat and Poultry Inspection program. Results from these three laboratories should be comparable to results that FSIS laboratories produce. Using a larger portion size increases the possibility of finding *Salmonella* in a given sample. Eight of the laboratories utilized methods that called for 25g sample portions, much less than the FSIS method requirement of a 325g portion. These methods are primarily used for outbreak sample analysis or samples already suspected of containing *Salmonella*.

Regardless of the method used for detecting *Salmonella*, laboratories were instructed to sample the food product following the FSIS MLG instruction for ready-to-eat foods. This requires taking the appropriate amount of sample by weight to include all ingredients without separating out the chicken portion of the product. Laboratories that performed aerobic count analysis followed their standard state protocol, as the timeframe of the project did not allow for all laboratories to become proficient in the FSIS aerobic count method.

Results and Discussion

Laboratories obtained samples at retail and provided results for 487 separate samples. It is important to note that these data were derived from a convenience sampling of eligible products available to the participating laboratories; no claims are made as to the statistical significance of any differences observed.

The overall positive rate for *Salmonella* in NRTE stuffed chicken products tested was 11.48% (Table 1). However, laboratories that used a sample size or methodology equal to FSIS reported 26.67% of samples positive for *Salmonella*. When considered by establishment, sample positive rates between 0 and 50% for *Salmonella* were observed (Table 2). Furthermore, when differing methodologies (*i.e.*, 25g vs. 325g product tested) were used to analyze samples from the same establishment, there was an increased percent positive rate associated with the larger analytical sample weight (Table 3). Results for aerobic count (AC) determinations are also shown for the eight laboratories that performed this analysis (Tables 1 and 2). While large variations in AC were observed, AC were similar between samples that were positive and negative for *Salmonella*, regardless of the sample size analyzed (Table 4).

Laboratories did not report any difficulty in sample preparation or analysis due to the breeding or different stuffing components. These results suggest that the FSIS sample size and sample preparation of including all ingredients (*i.e.*, without separating out the chicken portion of the product) is an acceptable testing method for this product.

Whole genome sequencing (WGS) was used to characterize the *Salmonella* isolates further. Analysts found *Salmonella* Enteritidis (18/58, 31%), *Salmonella* Infantis (22/58, 38%), *Salmonella* Kentucky (15/58, 26%) and *Salmonella* Typhimurium (3/58, 5%). These serotypes include those associated with the most recent NRTE stuffed chicken outbreaks and the most common serotypes associated with outbreak related illnesses^{2,3}. All WGS data is available in the National Center for Biotechnology Information (NCBI) database.

Highlights

- Participating laboratories using the same *Salmonella* detection and sample preparation as FSIS MLG 4.12 found *Salmonella* in these products nearly 27% of the time. This is similar to the rate of 29% found in further processed chicken parts or comminuted product, commonly used as the source material for this product¹.
- Using the larger sample size called for in the FSIS MLG 4.12 *Salmonella* method, laboratories detected *Salmonella* at a higher rate than in smaller size samples. This difference is likely due to low levels of *Salmonella* contamination or uneven distribution in the product.
- Whole genome sequencing (WGS) results identified all three of the FSIS Key Performance Indicator (KPI) serotypes Enteritidis, Infantis, and Typhimurium in these products. KPI strains represented 74% of all *Salmonella* positives; the remainder were *S.* Kentucky.

Table 1. *Salmonella* testing and aerobic counts in not Ready-to-Eat Breaded Stuffed Chicken Products

Laboratory	Samples (n)	Positives (n)	Positives (%)	Sample size analyzed (g)	AC (log) ± S.D.
CA	45	0	0.00	25	
CO	41	6	14.63	25	3.69 ± 0.61
FL	45	5	11.11	25	3.84 ± 0.51
MI	45	1	2.22	25	3.73 ± 0.59
MO	45	2	4.44	50	4.00 ± 0.56
NY	41	6	14.63	25	
TX	45	2	4.44	25	
VA	45	0	0.00	25	3.86 ± 0.61
Sub-total	352	22	6.25		
MN	45	12	26.67	375	3.81 ± 0.89
OH	45	11	24.44	325	3.77 ± 1.07
IA	45	13	28.89	325	4.05 ± 1.08
Sub-Total	135	36	26.67		
Overall Total	487	58	11.48		

Table 2. *Salmonella* testing and aerobic counts by establishment (blinded A-F) , with the number of labs involved in analyzing samples from each establishment

Establishment	Samples (n)	Positives (n)	Positives (%)	Labs (n)	AC (log) ± S.D.
A	46	7	15.22	4	4.02 ± 0.75
B	5	0	0.00	1	4.64 ± 1.55
C	72	10	13.89	5	3.79 ± 0.54
D	302	29	9.60	10	3.64 ± 0.62
E	12	6	50.00	5	6.43 ± 0.88
F	5	1	20.00	3	6.32 ± 0.57

Table 3. *Salmonella* results organized by establishment and further delineated by sample portion size analyzed.

Establishment	Sample Size Analyzed (g)	Positives (n)	Samples (n)	Positives (%)
A	25	2	31	6.4
A	325	2	5	40.0
A	375	3	10	30.0
B	325	0	5	0.0
C	25	0	11	0.0
C	50	1	15	6.7
C	325	1	21	4.7
C	375	8	25	32.0
D	25	12	229	5.2
D	50	1	15	6.7
D	325	15	50	30.0
D	375	1	8	12.5
E	25	1	4	25.0
E	325	5	6	83.0
E	375	0	2	0.0
F	25	0	2	0.0
F	325	1	3	33.3
*Unidentified	25	5	45	11.1
TOTALS		58	487	

*Unidentified - USDA Establishment information was not kept for these samples. However, they were identified as raw, breaded, stuffed chicken products.

Table 4. Log AC Counts vs positive and negative *Salmonella* samples

Sample Size analyzed (g)	AC results reported (n)	<i>Salmonella</i> Positives (n)	AC log (\pm SD) of Samples negative for <i>Salmonella</i>	AC log (\pm SD) of Samples positive for <i>Salmonella</i>
25	212	14	3.83 (0.58)	3.68 (0.42)
325	135	36	3.82 (1.08)	4.09 (1.03)

Table 5. *Salmonella* serotype and Whole Genome Sequencing (WGS) NCBI sample numbers for isolates, by product type and establishment.

Establishment	Sample Size Analyzed (g)	Positive <i>Salmonella</i> /Samples Tested	WGS NCBI Sample number	Serotype	Product Type
A	25	2/31	SAMN31282610 SAMN31660379	<i>S. Infantis</i> <i>S. Infantis</i>	Chicken Kiev Broccoli Cheese
A	325	2/5	SAMN31929773 SAMN31929778	<i>S. Infantis</i> <i>S. Enteritidis</i>	Broccoli Cheese Cordon Bleu
A	375	3/10	SAMN32041750 SAMN32388524 SAMN32041755	<i>S. Enteritidis</i> <i>S. Typhimurium</i> <i>S. Kentucky</i>	Cheese/ham Broccoli Cheese Swiss Cheese/Ham
B	325	0/5	N/A		
C	25	0/11	N/A		
C	50	1/15	SAMN31948492	<i>S. Enteritidis</i>	Chicken Kiev
C	325	1/21	SAMN31484236	<i>S. Enteritidis</i>	Ham/Cheese
C	375	8/25	SAMN31891079 SAMN32041754 SAMN32041757 SAMN32041752 SAMN32096665 SAMN32096652 SAMN32041751 SAMN32041756	<i>S. Enteritidis</i> <i>S. Kentucky</i> <i>S. Kentucky</i> <i>S. Kentucky</i> <i>S. Kentucky</i> <i>S. Kentucky</i> <i>S. Typhimurium</i> <i>S. Typhimurium</i>	Ham/Cheese Butter Blend Butter Blend Butter Blend Ham/Cheese Ham/Cheese Ham/Cheese Ham/Cheese
D	25	12/229	SAMN31370198 SAMN31370197 SAMN31370196 SAMN31370200 SAMN31370199 SAMN31370195 SAMN31660376 SAMN31660378 SAMN31660381 SAMN31660380 SAMN32258894 SAMN32259123	<i>S. Kentucky</i> <i>S. Infantis</i> <i>S. Infantis</i> <i>S. Infantis</i> <i>S. Enteritidis</i> <i>S. Enteritidis</i> <i>S. Infantis</i> <i>S. Infantis</i> <i>S. Infantis</i> <i>S. Enteritidis</i> <i>S. Enteritidis</i> <i>S. Infantis</i>	Broccoli Cheese Cordon Bleu Cordon Bleu Cordon Bleu Broccoli Cheese Broccoli Cheese Broccoli Cheese Broccoli Cheese Broccoli Cheese Broccoli Cheese Broccoli Cheese Cheeses and Ham
D	50	1/15	SAMN31948494	<i>S. Enteritidis</i>	Broccoli Cheese
D	325	15/50	SAMN31930292 SAMN31930295	<i>S. Kentucky</i> <i>S. Enteritidis</i>	Broccoli Cheese Cordon Bleu

			SAMN31930297 SAMN31930455 SAMN31929781 SAMN31929771 SAMN31929900 SAMN31484237 SAMN31484240 SAMN31484241 SAMN31484242 SAMN31484246 SAMN31537770 SAMN31930299 SAMN31929776	S. Infantis S. Kentucky S. Infantis S. Kentucky S. Enteritidis S. Enteritidis S. Enteritidis S. Enteritidis S. Kentucky S. Kentucky S. Enteritidis S. Infantis S. Infantis	Chicken Parm Broccoli Cheese Cordon Bleu Broccoli Cheese Crème Brie/Apple Broccoli Cheese Broccoli Cheese Ham/Cheese Asparagus/Cheese Broccoli Cheese Ham/Cheese Broccoli Cheese Cordon Bleu
D	375	1/8	SAMN32041753	S. enteritidis	Broccoli Cheese
E	25	1/4	SAMN31660377	S. Enteritidis	Ham/Swiss
E	325	5/6	SAMN31484238 SAMN31484239 SAMN31484243 SAMN31484244 SAMN31537769	S. Infantis S. Infantis S. Infantis S. Infantis S. Infantis	Spinach/artichk Ham/Swiss Spinach/artichk Ham/Swiss Spinach/artichk
E	375	0/2	N/A		
F	25	0/2	N/A		
F	325	1/3	SAMN31484245	S. Infantis	Broccoli/Cheese
N/A	25	5/45	SAMN32513378 SAMN32513376 SAMN32513375 SAMN32513374 SAMN32513377	S. Infantis S. Infantis S. Kentucky S. Kentucky S. Kentucky	Cordon Bleu Cordon Bleu Broccoli Cheese Broccoli Cheese Broccoli Cheese
TOTALS		58/487			

References

¹ https://www.fsis.usda.gov/sites/default/files/media_file/2022-02/FY2021-Sampling-Summary-Report.pdf

²FSIS Outbreak Reports at: <https://www.fsis.usda.gov/food-safety/foodborne-illness-and-disease/outbreaks>.

³ Centers for Disease Control and Prevention: National Outbreak Reporting System at: <https://wwwn.cdc.gov/norsdashboard/>.