Nationwide Young Chicken Microbiological Baseline Data Collection Program

November 1999 – October 2000

FOREWORD

This publication is a compilation of data obtained from the Nationwide Young Chicken Microbiological Baseline Data Collection Program for the twelve-month period of November 1999 through October 2000. The program was initiated by the Food Safety and Inspection Service (FSIS) to estimate the prevalence and levels of bacteria of public health concern on young chicken carcasses as currently produced. The program was designed through consultation with various staffs in the Agency and advice from scientists and organizations outside the Agency. The Microbiology Division (formerly the Biosciences Division) in conjunction with the Data Analysis and Statistical Support Staff (formerly the Evaluation and Analysis Division) coordinated the conduct of the program, provided data analysis and prepared this report. The microbiological analyses were conducted by the FSIS Field Service Laboratories located in Athens, GA, St. Louis, MO, and Alameda, CA. Sample collection was the responsibility of the FSIS Inspectors-in-Charge without whom this program could not have been accomplished.

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EXECUTIVE SUMMARY

From November 1999 through October 2000, rinse samples from 1,225 young chicken carcasses were collected by FSIS at establishments operating under Federal inspection. These samples were analyzed to estimate the prevalence and levels of selected bacteria of public health concern on young chicken carcasses as currently produced. The establishments sampled under the program are responsible for approximately 98% of all chickens slaughtered in the U.S. Young chicken carcasses were analyzed for the pathogen, *Salmonella*, and for generic *Escherichia coli*, an organism thought to be of value as an indicator of general hygiene and process control. Rinse samples from 1,225 chicken carcasses were analyzed quantitatively and qualitatively for generic *E. coli.* A qualitative analysis was performed for *Salmonella* spp. which were recovered from 8.7%; generic *E. coli* was recovered from 95.3%. Generic *E. coli* levels of 1,000 or fewer colony forming units per milliliter (cfu/ml) were found in 99.1% of the rinse samples positive for *E. coli*.

INTRODUCTION

The Food Safety and Inspection Service (FSIS) is the Federal agency responsible for enforcing the Federal Meat Inspection Act, the Poultry Products Inspection Act and the Egg Products Inspection Act. These Acts empower the Agency to review facilities for evidence of insanitation, to inspect final products for evidence of adulteration and to review labels to ensure proper product labeling. The Acts stipulate the penalties that the Agency can impose to ensure compliance. The inspection of food animals at the time of slaughter has historically focused on identifying symptoms of disease conditions that make the animal carcass or parts of the carcass unfit for human food. Many human pathogens, however, reside harmlessly on the hide, feathers or skin of healthy animals or in their digestive tracts, causing no symptoms of disease. Bacteria of many types are normal and unavoidable residents of all warm-blooded animals, including humans. Bacteria are not detectable by visual inspection. The slaughter procedures that have developed over the years, as well as antimicrobial interventions recently incorporated by industry (e.g. trisodium phosphate washes, organic acid rinses, steam vacuuming, steam pasteurization), reduce the levels of many human pathogens but do not completely eliminate them. The production of raw meat and poultry does not include a lethal processing intervention, such as a cook step, that is designed to kill remaining bacteria. Therefore, even when produced under ideal conditions, carcasses from

normal, healthy young chickens can potentially contain a variety of bacteria, including spoilage organisms and low levels of some pathogens. This fact has long been recognized by the Agency and by scientific experts around the world. Therefore, if poultry is not properly cooked, held, cooled, and stored, there is the potential for any pathogens present to proliferate and cause foodborne illness in humans.

OBJECTIVES

This non-regulatory program had three primary objectives[1]:

1. To collect nationwide data that provide a general microbiological profile of young chickens for selected microorganisms of various degrees of public health concern.

2. To compare data on *Salmonella* and generic *E. coli* populations with findings from the earlier baseline study [2].

3. To use the information and knowledge gained from this program as a reference for further investigations and evaluation of new prevention programs.

Program Design Relative to Objectives:

This *Nationwide Young Chicken Microbiological Baseline Data Collection Program* establishes an updated profile of *Salmonella* and generic *E. coli* in young chickens as currently produced under Federal inspection. The results on the presence and quantity of selected microorganisms are expressed as a national average relative to slaughter volume. This approach is similar to the earlier FSIS Nationwide Microbiological Baseline Data Collection Programs for broiler chickens[2], steers and heifers[3], cows and bulls[4], market hogs[5], and young turkeys[6].

PROGRAM DESIGN

Establishments Included in the Sampling Frame:

All establishments that consistently slaughtered young chickens under Federal inspection during the sample collection period were included in the sampling frame. There were approximately 205 establishments in this category. Young chicken production accounted for approximately 98% of all chicken production within Federally inspected plants.

Sample Design:

Many factors were considered in the design of this sampling program. Among these were the size and variability of the young chicken population, the nature and number of bacteria to be investigated, the practical costs of sampling, competing program demands, and the type of information sought.

Both sampling and non-sampling errors can affect the reliability of results and, thus, had to be considered in designing this program. Sampling errors occur because observations are derived from a portion rather than from the entire population; non-sampling errors can be attributed to many sources inherent in the collection of samples, laboratory analysis and processing of data. Both types of errors were considered in determining the sample size.

In order to approximate a random sample, the selection was performed in two stages. The first stage was to randomly select a young chicken establishment from the sampling frame, and the second was to randomly select the young chicken carcass sample. Establishments were selected with probabilities assigned in proportion to their total number of birds slaughtered. Establishments that produced a larger percentage of the total number of birds slaughtered had a greater chance of being selected than smaller establishments, and could be selected more than once. This ensured that all young chickens slaughtered had an approximately equal chance of being selected. The program was not designed to provide microbiological information on individual establishments. In order to obtain such information, one would need to collect a large number of samples from each establishment over a period of time. Also, the *Salmonella* results obtained from this one-year program were independent from the Agency's ongoing *Salmonella* Hazard Analysis and Critical Control Point (HACCP) testing program.

It was determined that about 1,200 samples would ensure reasonable levels of precision for yearly estimates given the expected prevalence for the bacteria included in this study. To achieve this number, a random sample of 1872 young chicken carcasses was requested during the 52-week time frame of the study (approximately 36 per week).

Sampling Location within the Establishment:

In order to accomplish the objectives of this program, it was imperative to acquire data from a significant point in the production process. Key factors in the microbial profile of young chickens are the slaughter and evisceration processes conducted under Federal inspection. To evaluate the cumulative effects of these processes, carcasses were sampled from the drip line after the chill tank. This is the endpoint of slaughter and evisceration, preceding any additional processing, which would introduce variables that could interfere with the interpretation of the data intended to describe slaughter and evisceration processes.

Sample Collection and Description:

Samples were aseptically collected by FSIS Inspectors-in-Charge following the procedures in FSIS Directive 10,230.5 (2/4/98), instructions provided on computer-generated sample collection request forms, and specific instructions applicable to this program. For each sample, one randomly selected post-chiller whole young chicken carcass was aseptically placed into a sterile bag and shaken with 400 ml prechilled Buffered Peptone Water (BPW). Once the contents of the bag were properly mixed, two sterile screw-cap containers with lids were each filled with 100 ml of rinse fluid, i.e. a total of 200 ml rinse was submitted for analysis. Portions of the carcass rinse were used for each of the ensuing microbiological analyses. The two sample containers were sealed, put into individual resealable bags, placed in an insulated shipper with gel packs capable of maintaining refrigeration temperatures, and shipped to the designated laboratory by an overnight delivery service. Rinse samples from these carcasses were collected Monday through Friday during slaughter operations and were shipped chilled to the laboratory by an overnight courier on the same calendar day they were collected. Only those samples received at the laboratory the day after sample collection, with a sample receipt temperature of 0°C to 10°C (inclusive) were analyzed. Samples received outside this temperature range were not analyzed.

Selection of Organisms:

A discussion of the choice of organisms to be used in establishing microbiological guidelines is found in the study entitled "An Evaluation of the Role of Microbiological Criteria for Foods and Food Ingredients" published by the Subcommittee on Microbiological Criteria for Foods and Food Ingredients of the National Research Council, National Academy of Sciences[7] . That rationale was used in previous FSIS baseline sampling programs and was reviewed and assessed for incorporation into this program.

For the purposes of the Young Chicken Microbiological Baseline Data Collection Program, two microorganisms were selected for analysis: *Salmonella* (non typhi) was selected from organisms most often associated with human illness as determined by foodborne illness reports [8, 9, 10]. A second organism was chosen as an indicator of general hygiene and process control (generic *Escherichia coli*).

Analytical Methods:

Samples were analyzed according to the procedure for quantitative generic *E. coli* analysis described in Chapter 3, Section 3.5c of the USDA/FSIS *Microbiology Laboratory Guidebook* (*MLG*) (3rd ed., 1998)[11] employing rehydratable Petrifilm™ to obtain generic *E. coli* counts. Appropriate dilutions of 10^{-1} to 10^{-3} of carcass rinse fluid were made to obtain an end point. Petrifilm™ was inoculated in duplicate using 1.0 ml of the undiluted carcass rinse fluid and 1.0 ml of each dilution. The Petrifilm™ was then incubated and resultant colonies counted as per manufacturer's directions. Generic *E. coli* counts were reported as cfu/ml of carcass rinse fluid analyzed.

For *Salmonella* analysis, 30 ml of sterile single-strength BPW was added to 30 ml of carcass rinse fluid from the sample container to bring the total volume to 60 ml. Procedures for the analysis of qualitative *Salmonella* were followed according to the instructions in Chapter 4 of the *MLG*[12], beginning at Section 4.45c.

RESULTS

Data were obtained for 1,225 young chicken carcasses. Some samples were not collected for various reasons, e.g. the establishment did not slaughter that particular week. Other samples were collected but not analyzed if, for example, they became compromised during shipment (e.g., open package, invalid temperature, delayed shipment, etc.). Table 1 presents the prevalence, or frequency of occurrence, of the selected microorganisms in tested portions of the young chicken carcass rinse fluids. An estimated national prevalence of 95.3% for generic *E. coli* was observed in the 1,225 young chicken carcass rinse samples analyzed. The prevalence estimate for *Salmonella* was 8.7%.

Table 2 presents the mean level of generic *E. coli* recovered from the young chicken carcass rinses that tested positive. Due to the broad range of quantitative values observed, mean levels in Table 2 are expressed as both the $\log_{10}$ mean and the geometric mean; the geometric mean is the antilog of the $\log_{10}$ mean. Table 3 shows the frequency within which all samples enumerated for generic *E. coli* fall within specified intervals. Young chicken carcass rinse fluids that tested positive for generic *E. coli* had a geometric mean of viable organisms of 12.10 (cfu/ml) (Table 2) with 99.1% of carcass rinse fluids containing 1,000 or fewer generic *E. coli* per ml (Table 3, Figure 1).

DISCUSSION

The program was designed to provide estimates of national prevalence and levels of selected microorganisms on young chicken carcasses produced in Federally inspected plants. In 1994 - 1995, FSIS conducted a similar baseline study for *Salmonella* and Biotype I (generic) *E. coli* (National Broiler Chicken Microbiological Baseline Data Collection Program-April, 1996) [2]. The 1994 - 1995 study also included testing for *Listeria monocytogenes*, *E. coli* O157:H7, aerobic plate count, *Staphylococcus aureus*, coliforms and *Clostridium perfringens*. Because FSIS had discontinued sub-classification of broilers among the slightly larger population of “young chickens”, the 1999-2000 protocol specified sampling of the latter. Chicken carcasses were submitted for testing during the 1994-1995 study, while only carcass rinses were submitted for testing in the 1999-2000 study. No appreciable difference in pathogen numbers have

been found when BPW carcass rinses are held cold for 24 hours and then analyzed vs. chicken carcasses that were rinsed immediately prior to analysis.

This report presents the primary findings of this program: an updated microbial profile of young chicken carcasses, as produced under Federal inspection, stating the prevalence of *Salmonella* and the prevalence and level of generic *E. coli* as determined from carcass rinse samples. This baseline study found prevalences of 8.7% for *Salmonella* and 95.3% for generic *E. coli* with a mean number of 12.10 cfu generic *E. coli* per ml of carcass rinse fluid.

The potential presence of pathogenic bacteria on young chicken carcasses emphasizes the need for proper refrigeration, handling and cooking of chicken products throughout the food chain. Based on these, and previous baseline data, it appears that chicken products cooked to achieve a 6.5 $\log_{10}$ decrease in salmonellae, as expressed in an FSIS publication on performance standards for certain meat and poultry products[13], should not contain viable *Salmonella* after cooking. These results also underscore the need for special care so that cross contamination of ready-to-eat food products can be avoided, and also for thorough cleaning and disinfection of food preparation work surfaces and utensils after handling raw chicken products.

TABLES

Table 1. Prevalence of Selected Microorganisms in Tested Portions (*n*=1225) of Young Chicken Carcass Rinse Fluids

Microorganism	Number Positive	Prevalence (%)	SE[1]
INDICATOR ORGANISM			
Generic *Escherichia coli*	1167	95.3	0.6
PATHOGENIC ORGANISM			
Salmonella (non typhi)	107	8.7	0.8

[1] Standard error of prevalence using binomial distribution.

Source: USDA/FSIS Nationwide Young Chicken Microbiological Baseline Data Collection Program: November 1999 - October 2000.

Table 2. Mean Level of Generic *Escherichia coli*[1] in Young Chicken Carcass Rinse Fluids (*n*= 1225)

Microorganism	No. of positive samples	Mean Populations[2]			
		Log_{10}		Geometric Mean	
		Mean	SE[3]	Mean	95% CI[4]
Generic *E. coli*	1167	1.08	0.02	12.10	11.13, 13.15

[1]Expressed as cfu/ml.

[2]Mean includes positive samples only.

[3]Standard error of the mean log of positive samples.

[4]Confidence interval.

Source: USDA/FSIS Nationwide Young Chicken Microbiological Baseline Data Collection Program: November 1999 - October 2000.

Table 3. Generic *Escherichia coli* Distribution in Young Chicken Carcass Rinse Fluids

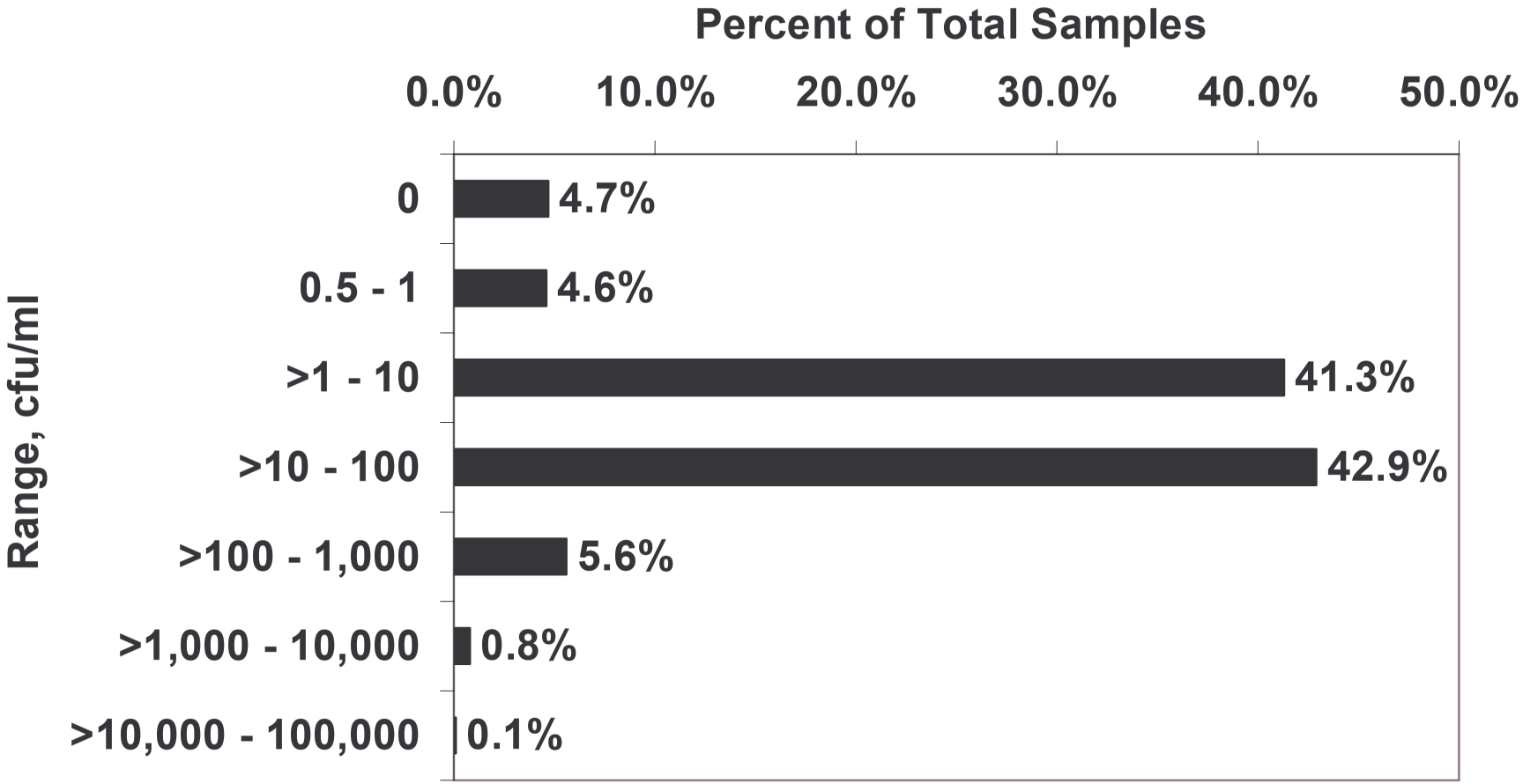
Range (cfu/ml)	No. of samples	Percent of total	Cumulative no.	Cumulative percent
0[1]	58	4.7	58	4.7
0.5-1	56	4.6	114	9.3
>1-10	506	41.3	620	50.6
>10-100	526	42.9	1146	93.6
>100-1,000	68	5.6	1214	99.1
>1,000-10,000	10	0.8	1224	99.9
>10,000-100,000	1	0.1	1225	100.0
TOTALS	1225	100.0		

[1]Negative by test method (lower limit of detection is 0.5 cfu/ml).

Source: USDA/FSIS Nationwide Young Chicken Microbiological Baseline Data Collection Program: November 1999 - October 2000.

FIGURE

Figure 1. Ranges of Generic *Escherichia coli* Distribution (cfu/ml) in Young Chicken Carcass Rinse Fluids



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