Impact of Feathers and Feather Follicles on Broiler Carcass Bacteria

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ABSTRACT Genetically featherless and feathered broiler siblings were used to test the contribution of feathers and feather follicles to the numbers of aerobic bacteria, *Escherichia coli*, and *Campylobacter* in whole-carcass rinse samples taken immediately after carcasses were defeathered for 30 or 60 s. Numbers of spoilage bacteria were counted after the same fully processed carcasses were stored for 1 wk at 2°C. In each of 3 replications, twenty-eight 11-wk-old, mixed-sex, genetically featherless or feathered broilers were processed in a laboratory processing facility. Immediately after individual defeathering in a mechanical picker, carcasses were sampled using a carcass rinse technique. Carcasses were eviscerated, immersion chilled at 2°C for 30 min, individually bagged, and stored for 1 wk at 2°C, after which all carcasses were rinsed again, and spoilage bacteria in the rinsate were enumerated. There were no significant differences ($P \leq 0.05$) between the featherless and feathered broilers in numbers of aerobic bacteria, *E. coli*, and *Campylobacter* in rinse samples taken immediately after defeathering and no differences between carcasses picked for 30 or 60 s. There were no differences in numbers of spoilage bacteria after 1 wk of refrigeration for any of the feather presence-picking length combinations. Although the defeathering step in poultry processing has been identified as an opportunity for bacterial contamination from the intestinal tract and cross-contamination between carcasses, the presence of feathers and feather follicles does not make a significant difference in carcass bacterial contamination immediately after defeathering or in spoilage bacteria after 1 wk of refrigeration.

(*Key words*: aerobic bacteria, *Campylobacter*, *Escherichia coli*, feather follicle, skin)

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INTRODUCTION

The importance of bacteria in feather follicles seems to be a firmly established concept (International Commission on Microbiological Specifications for Foods, 1980; National Advisory Committee on Microbiological Criteria for Foods, 1997) for pathogenic and spoilage bacteria. McMeeken and Thomas (1979) observed, however, that there was little experimental evidence of the location of bacteria in follicles except for in tetrazolium dye studies (Barnes and Impey, 1968; Barnes et al., 1973). Berndtson et al. (1992) found *Campylobacter* in scrapings of empty feather follicles of chilled carcasses, but similar samples were not taken before processing. Kim et al. (1996) photographed *Salmonella* in feather follicles of skin soaked for 2 h in a highly concentrated suspension of *Salmonella*, but those conditions were extreme compared with what carcasses experience during commercial processing.

The growth of bacteria in feather follicles rather than on the skin surface has been suggested by Barnes et al. (1973) as the reason that maceration of poultry skin samples yielded higher numbers of bacteria in the work of Avens and Miller (1973). Avens and Miller, however, found that the number of subcutaneous bacteria was not large enough to have a significant influence on the number of bacteria that could be recovered. In a direct test of the contribution of follicles to total carcass bacteria, Buhr et al. (2003) demonstrated that there was little or no difference in bacterial counts of stomached breast skin from genetically featherless (and thus without follicles) and normally feathered broilers when skin samples were taken immediately after carcasses passed through defeathering machines. Considering the scale of carcass and skin topography and the relative size of bacteria, there are many sites on a carcass for bacteria to attach or become entrapped without relying on feather follicles.

The possible role of feathers in bacterial contamination of carcasses is not limited to the follicles alone. The feathers themselves carry a large bacterial population on arrival at the processing plant (Kotula and Pandya, 1995; Georgnaras et al., 1997). The defeathering process clearly contributes to bacterial contamination and cross-contamination (Mulder et al., 1978; Oosterom et al., 1983; Berrang et al., 2001; Allen et al., 2003), although Buhr et al. (2000) demonstrated that groups of carcasses arriving for processing with significantly different numbers of external

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bacteria can be microbiologically indistinguishable in samples taken after scalding and defeathering.

Because of the necessity of removing all feathers, most processing plants have excess defeathering capacity. Allen et al. (2003) reported that the majority of feathers were removed in the first 10 s in the defeathering machines, which is consistent with our own observations, even though carcasses are defeathered for 30 s or more in most processing plants. The excess defeathering capacity provides insurance against missed feathers if some problem arises, but there is a possibility of increased cross-contamination and skin damage. Several studies have reported that fecal bacteria escape from the intestinal tract (Oosterom et al., 1983; Berrang et al., 2001), possibly because of pressure from the defeathering machines; thus any unnecessary defeathering may increase the possibility of fecal contamination. The present experiment was carried out to investigate the role played by feathers and feather follicles in bacterial contamination of carcasses during processing. The time length of defeathering was also varied (30 and 60 s) to test for any effect on numbers of carcass bacteria.

MATERIALS AND METHODS

Featherless and Feathered Broilers

The origin and husbandry of the genetically featherless and feathered broiler siblings used in this experiment were described by Buhr et al. (2003). That paper included photographs of both types of carcasses after defeathering, showing the lack of feather follicles on the skin of the featherless chickens. Featherless and feathered, mixed-sex siblings were reared to 11 wk of age on pine shavings in floor pens. To equalize the effects of any pressure on the carcasses during defeathering, birds to be processed were selected 1 wk before the experiment to provide approximately the same mean body weight in the featherless and feathered broilers. One week before processing, all birds were challenged orally with 2 mL of approximately 10^8 cfu/mL of *Campylobacter jejuni* Strain PPMQ2b, which was obtained from whole-carcass rinses of processed broiler carcasses at a local poultry processing facility. Serial dilutions of carcass rinsates were plated on Bacto *Campylobacter* agar kit blaser,[2] and plates were incubated in a BBL GasPak jar[3] with an activated CampyPak Plus hydrogen + CO_2 with integral palladium catalyst[3] for 48 h at 42°C. *Campylobacter*-like colonies were removed from the incubated plates and presumptively identified using the latex-Campy (jcl) *Campylobacter* culture confirmation test.[4] Isolates that were positive on the latex agglutination test were confirmed as *Campylobacter jejuni* using the MIDI Sherlock Microbial Identification System.[5] Stock cultures of the isolate were maintained by transferring to fresh blood agar plates[6] at weekly intervals and incubating in a BBL GasPak with a CampyPak Plus hydrogen + CO_2 with integral palladium catalyst for 48 h at 37°C. Stock cultures were stored at 4°C, and fresh *C. jejuni* cultures were prepared by streaking stock cultures onto blood agar plates and incubating in a BBL GasPak Jar with a CampyPak plus hydrogen + CO_2 with integral palladium catalyst for 48 h at 37°C. After incubation, cultures were harvested by adding 9 mL of a 0.1% Bacto peptone[2] solution to the surface of the plates and using sterile bacterial cell spreaders to scrape cell growth from the plates. Harvested bacterial suspensions contained approximately 10^8 cfu/mL.

Processing

Three replications of the experiment were conducted with 28 birds in each. Feed withdrawal was accomplished by catching the broilers the night before processing and placing them in plastic coops, where they remained until processing the following morning, approximately 12 h later. Four treatment combinations (2 feather conditions by 2 defeathering times) were alternated during processing. Chickens were removed individually from coops, wing-banded, and placed in shackles. They were electrically stunned (50 V AC) for 10 s, then bled for 90 s, and scalded for 90 s at 54°C. Individual carcasses were mechanically defeathered in a commercial 5-bank feather picker[7] for 30 or 60 s by changing the line speed. Rubber fingers in the defeathering machines were sprayed clean with hot water (approximately 82°C) between carcasses. Ability of spray cleaning with hot water to reduce cross-contamination by *Campylobacter* and other bacteria has been shown in earlier experiments (Musgrove et al., 1997).

After removal of necks and feet, defeathered carcasses were placed in new plastic bags, and 200 mL of 0.1% peptone water[2] was added. Carcasses were placed in an automated shaker for 1 min (Dickens et al., 1985), after which carcasses were removed from the bags. The rinse liquid was poured into sterile plastic containers that were refrigerated at 4°C until microbiological analysis was performed (within 1 h).

After the rinse sampling, carcasses were eviscerated by hand and batch chilled for 30 min in a mixture of ice and water in a paddle-type chiller turning at 4 rpm to simulate tumbling in commercial chillers. After being chilled, carcasses were drained briefly and placed into clean plastic bags. After 1 wk in a refrigerator at 2°C, carcasses were removed, placed in fresh plastic bags with 200 mL of 0.1% peptone water, and rinsed for 1 min in an automated shaker.

Microbiological Analysis

Whole-carcass rinse samples taken immediately after chilling were cultured for aerobic bacteria, *E. coli*, and *Campylobacter*. Rinse samples taken after 1 wk of storage

[2]Becton Dickinson Microbiology Systems, Sparks, MD.
[4]MIDI, Inc., Newark, DE.
[5]Remel, Lenexa, KS.
[6]Difco Laboratories, Detroit, MI.
[7]Johnson Food Equipment Co., Kansas City, KS.
[3]PanBio, Inc., Baltimore, MD.

TABLE 1. Mean $\log_{10}$ (cfu/mL of rinse) for aerobic bacteria, *Escherichia coli*, and *Campylobacter* in carcass rinses immediately after defeathering and spoilage bacteria in carcass rinses after holding eviscerated, immersion-chilled carcasses for 1 wk at refrigerator temperature[1]

		Defeathering time (s)		
	Type	30 s	60 s	Mean
Aerobic bacteria	Featherless	4.0 ± 0.5	3.9 ± 0.4	3.9 ± 0.4
	Feathered	3.9 ± 0.5	3.9 ± 0.7	3.9 ± 0.6
	Mean	4.0 ± 0.5	3.9 ± 0.6	
E. coli	Featherless	2.9 ± 1.0	2.8 ± 0.8	2.8 ± 0.9
	Feathered	3.0 ± 0.8	3.0 ± 0.8	3.0 ± 0.8
	Mean	2.9 ± 0.9	2.9 ± 0.8	
Campylobacter	Featherless	2.9 ± 0.9	2.5 ± 1.0	2.7 ± 0.9
	Feathered	2.4 ± 0.6	2.6 ± 1.2	2.5 ± 0.9
	Mean	2.6 ± 0.7	2.5 ± 1.1	
Spoilage	Featherless	2.2 ± 0.8	3.0 ± 1.3	2.3 ± 1.1
	Feathered	2.6 ± 0.9	2.8 ± 1.1	2.7 ± 1.0
	Mean	2.4 ± 0.8	2.7 ± 1.2	

[1]Least square means from 3 replicates ± SD (n = 21) for each feather type and defeathering time combination.

were cultured for spoilage bacteria. Aerobic bacteria were enumerated by making a series of 1:10 dilutions and plating on plate count agar[2] that was incubated at 35°C for 48 h. *E. coli* bacteria were enumerated by adding 1 mL of serial dilutions of the rinsate to Petrifilm,[8] which was incubated as suggested by the manufacturer. Serial dilutions were also plated on Bacto *Campylobacter* agar, blaser.[2] Inoculated plates were incubated for 48 h at 42°C in a BBL GasPak jar system[7] containing an activated BBL CampyPak Plus gas generator envelope. After incubation, *Campylobacter*-like colony-forming units were counted. Selected colonies were subjected to a latex-*Campy* (jcl) *Campylobacter* culture confirmation test[9] to confirm identity as *Campylobacter* spp. Spoilage bacteria were enumerated by plating serial dilutions on plate count agar that was incubated at 4°C for 10 d before counting of colonies.

Statistical Analysis

Four of the 84 carcasses were *Campylobacter* negative, and those data were treated as missing values in the *Campylobacter* analysis. Other bacterial counts were transformed to $\log_{10}$ (cfu/mL) before statistical analysis with PROC general linear model of SAS software (SAS Institute, 1987). The data were analyzed in a 2-by-2 factorial arrangement with interaction terms and with statistical significance determined at $P \leq 0.05$.

RESULTS AND DISCUSSION

Effect of Feathers and Feather Follicles

Results for genetically featherless and feathered carcasses for aerobic bacteria, *E. coli*, and *Campylobacter* are shown in Table 1. There were no differences in bacterial counts between featherless and feathered carcasses in carcass rinses samples taken immediately after defeathering. Buhr et al. (2003) also found no differences in aerobic bacteria and *E. coli* on stomached breast skin samples from featherless and feathered carcasses after defeathering when the vents were untreated as in normal commercial processing and in 1 of 2 trials when the vents were plugged with tampons and sewn shut to block escape of fecal bacteria. There were significant differences in *Campylobacter* counts reported by Buhr et al. (2003), however, when vents were untreated, and higher numbers were on feathered carcasses. It is not clear whether the type of sample (breast skin samples vs. whole-carcass rinses) might have influenced the difference in *Campylobacter* results between the 2 studies.

Means of spoilage bacteria are also shown in Table 1. During chilling and refrigerated storage for 1 wk, there were no differences in numbers of spoilage bacteria recovered from rinses of carcasses that were featherless (no follicles) or feathered (with follicles) before picking. Barnes and Impey (1968) reported that when carcasses were stored at 1°C until a slight off-odor developed, reduction of tetrazolium showed that bacteria were growing in empty feather follicles. The numbers of spoilage bacteria in feather follicles must not be high enough to increase the total number of spoilage bacteria that can be recovered in carcass rinses of carcasses that have follicles, relative to carcasses without follicles. There are many sites on a carcass for bacteria to attach or become entrapped (McMeeken and Thomas, 1979), even on genetically featherless broilers without feather follicles.

Effect of Defeathering Times

As shown in Table 1, there were no significant differences in bacterial counts between 30 and 60 s defeathering times for aerobic bacteria, *E. coli*, and *Campylobacter*. Rathgeber and Waldroup (1994) reported that repicking of eviscerated, chilled half-carcasses reduced numbers of aerobic bacteria by approximately 0.8 logs and *E. coli* by approximately 1.9 logs on the half carcasses that were

[8]3M Health Care, St. Paul, MN.
[9]Integrated Diagnostics, Baltimore MD.

picked a second time. Those results were not repeated in the present experiment with unevisceratated, unchilled whole carcasses. Slavik et al. (1995) found significant differences in most probable numbers of inoculated *Salmonella* on chilled carcasses in 2 of 3 trials when carcasses were defeathered for 60 or 78 s, but the defeathering times in that experiment differed by approximately 30% (vs. 100% in the present experiment) and were confounded with 1- or 2-min scalding times and 60 or 52°C scald water, respectively. It might be possible for different defeathering times to make a difference in carcass bacteria if the combination of scalding temperature and defeathering time made a difference in whether the cuticle or outer layers of the epidermis was being removed by the longer defeathering time with a possible change in the number of skin bacteria that might be removed. Allen et al. (2003) also noted that the degree of bacterial dispersion and cross-contamination was influenced by the design of the defeathering equipment and the direction in which rubber fingers were rotating.

No significant difference was found in counts of spoilage bacteria between 30 and 60 s defeathering times (Table 1). Damage to skin caused by the defeathering machines could possibly create a more favorable environment for growth of spoilage bacteria. Fletcher and Thomason (1980) reported that longer defeathering times increased the incidence of oily bird syndrome. In the present experiment, some of the carcasses defeathered for 60 s were greasy to the touch, but counts of bacteria were not affected.

The presence of feathers makes defeathering a necessary step in poultry processing, even though defeathering is clearly involved in bacterial contamination and cross-contamination of carcasses (Mulder et al., 1978; Oosterom et al., 1983; Berrang et al., 2001; Allen et al., 2003). Feathers are highly contaminated when poultry arrive at the processing plant (Kotula and Pandya, 1995; Georgnaras et al., 1997), although scalding and defeathering can remove a large proportion of any external contamination (Buhr et al., 2000). The work of Buhr et al. (2003) and the results of the present experiment indicate that feather follicles make only a minor contribution to carcass bacteria.

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