Campylobacter jejuni Seasonal Recovery Observations of Retail Market Broilers[1]

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ABSTRACT This study investigated possible seasonal trends in the *Campylobacter jejuni* carrier state of market broilers. In this study, broiler carcasses, 15 each of two major companies, were obtained from a local supermarket each month for an entire year to evaluate the presence of *C. jejuni* on the carcasses. Direct plating and the whole carcass rinse procedure were used for *C. jejuni* detection. Resuscitation of damaged cells and preenrichment of low numbers of microorganisms were accomplished by Hunt's procedure. None of the carcasses tested positive from direct plating of skin flora in this study. After both Company A and Company B broiler samples were enriched, 69% (229/330) of the raw commercial broilers were positive for *C. jejuni*. The highest recovery rates were obtained during the warmer months of the year, from May through October (93, 97, 97, 87, 87, and 93% respectively), and the lowest were obtained in December (7%) and January (33%). Storage time, due to slow movement of broilers, appeared to affect the detectability of *C. jejuni* during December and January. This study shows that seasons of the year influence *C. jejuni* detectability and the carrier state in market broilers at retail level.

(*Key words*: *Campylobacter jejuni*, broiler, seasonal variation, colonization, observations)

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INTRODUCTION

Increased public concern over foodborne diseases associated with poultry has highlighted the need to monitor and improve control over the presence of microorganisms in the poultry marketing chain. Past studies have demonstrated that *Campylobacter jejuni* is typically found in large numbers and in a high percentage of broilers (Smitherman *et al*., 1984; Genigeorgis *et al*., 1986). *Campylobacter jejuni* is commonly found in the intestinal tract of chickens and is transferred to the skin during slaughter and processing. Jones *et al*. (1991) reported that 20% of cloacal samples were positive for *C. jejuni* as birds entered a processing plant, 52% following chilling and 31.6% at the retail level. The prevalence of *C. jejuni* varies in flocks, with some flocks escaping infection at the production level (Annan-Brah and Janc, 1988; Willis *et al*., 1991). The colonization rate of *C. jejuni* in poultry has been found to vary from 0 to 100% (Acuff *et al*., 1982; Genigeorgis *et al*., 1986; Jones *et al*., 1991; Willis *et al*., 1991). Generally, more flocks remain free of this infection during the cooler months of the year (Willis *et al*., 1991).

Campylobacter jejuni infection in humans has shown peak isolation rates during the summer (Tauxe, 1992). Conversely, a study conducted by Mattila *et al*. (1992) in Finland found that *Campylobacter* strains were the leading cause of travelers' diarrhea in the winter (28%), and caused only 7% of these cases in the fall. Substantial variability exists in the colonization of *C. jejuni* in different broiler flocks, and at different ages in the production cycle. The incidence of colonization increases with age.

In response to the Stern (1995) report on the influence of season and refrigerated storage on *Campylobacter* spp. contamination of broiler carcasses, and considering the lack of published information on seasonal variability in broiler flocks and retail market broilers in the U.S., the following study was conducted to determine whether trends relating to *C. jejuni* seasonal detectability exist in retail market broilers.

MATERIALS AND METHODS

The retail-level market broiler chickens of two major poultry integrated companies were used to assess the carrier state for *C. jejuni*. This study was conducted over a one-year period beginning in the fall and ending in the summer. A total of 30 whole broiler carcasses (15 from each company) were obtained monthly for 1 yr. Skin swab samples from the broilers were taken and plated onto Campy-Cefex medium (Stern *et al*., 1992) for

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TABLE 1. Recovery of *Campylobacter jejuni* from two brand-named retail-level chicken carcasses

Month	Company	Positive samples per total samples	Percentage positive samples	Overall totals[1]
January	Company A	9/15	60.0	10/30 33.3
	Company B	1/15	6.7	
February	Company A	1/15	6.7	14/30 46.7
	Company B	13/15	86.7	
March[2]	Company A	0/15	0	0/30 0
	Company B	0/15	0	
April	Company A	2/15	13.3	16/30 53.3
	Company B	14/15	93.3	
May	Company A	14/15	93.3	28/30 93.3
	Company B	14/15	93.3	
June	Company A	15/15	100	29/30 96.7
	Company B	14/15	93.3	
July	Company A	15/15	100	29/30 96.7
	Company B	14/15	93.3	
August	Company A	13/15	86.7	26/30 86.7
	Company B	13/15	86.7	
September	Company A	11/15	73.3	26/30 86.7
	Company B	15/15	100	
October	Company A	13/15	86.7	28/30 93.3
	Company B	15/15	100	
November	Company A	15/15	100	21/30 70.0
	Company B	6/15	40	
December	Company A	0/15	0	2/30 6.7
	Company B	2/15	13.3	

[2]Power outage in building during this month while samples were in water bath.

[1]Campylobacter-positive samples/total samples tested (percentage positive).

detection assessment. The whole carcasses were manually washed by shaking them in a large plastic bag containing 200 mL of sterile buffered peptone broth to obtain carcass-associated microflora. The suspensions were filtered into sterile 50-mL centrifuge tubes. The washed suspension was centrifuged for 10 min at 8,000 × *g*. The supernatant was removed and pellets were plated onto Campy-Cefex medium. The remaining pellets were resuspended in 5 mL of buffered peptone, then 1 mL of the resuspended mixture was added to 100 mL of Hunt's enrichment broth (Hunt, 1992). The broth mixture was incubated in a water bath for 24 h at 32 C for 3 h, then 37 C for 2 h, and 42 C for the remaining time. Samples then were directly plated onto Campy-Cefex medium, placed in air-locked baggies filled with a gas mixture (5% 0_2, 10% CO_2 and 85% N_2) and incubated for 24 h at 42 C. The plates were then observed for *C. jejuni*. Confirmation was made by microscopic observation and other biochemical tests (Gram-stain, catalase, oxidase, and hydrolyzed hippurate). Samples were enumerated as positives and percentage positives.

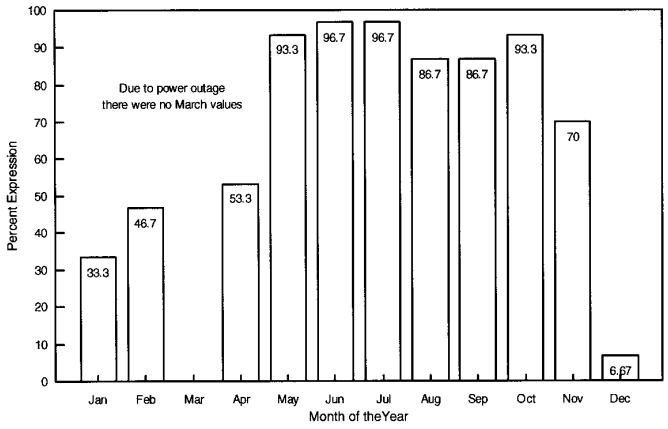

FIGURE 1. Seasonal percentages of *Campylobacter jejuni* positive carcasses of retail broilers in a 1-yr study.

RESULTS AND DISCUSSION

Recovery of *C. jejuni* from chicken carcasses by direct plating of samples was not effective in this experiment. None of the carcasses tested positive by direct streaking of skin carcass flora onto plating media during the 12 mo of observation. To enhance detectability, all samples required the utilization of an enrichment broth.

The monthly percentages of positive carcass samples using an enrichment broth are shown in Table 1. Of the 330 broilers sampled, 229 tested positive (69.4%) for *C. jejuni*. Of the total number (360), 30 carcass samples are not included due to an overnight power failure in March. When results from the two companies were compared, no major difference was observed. Company A had 65% (108/165) testing positive for *C. jejuni*, and Company B had 73% (121 of 165) positive.

It appears that a seasonal variation of detection rates of *C. jejuni* existed in these retail broiler carcasses. Figure 1 shows the percentages of *Campylobacter*-positive broiler carcasses per sampling month. The figure shows a seasonal pattern in the *C. jejuni* contamination detection rate of broiler carcasses, with highest detection rates between May and October (93, 97, 97, 87, 87, and 93% respectively), and the lowest in December (6.7%) and January (33%).

The increase in the number of positive specimens is similar to those observed for farm-raised broilers in cages and on floors (Willis *et al*., 1991) during the warmer months. In many cases, *C. jejuni* could not be detected during the winter months, as is described in subsequent studies.

Numerous other publications (Park *et al*., 1981, Aho and Hirn, 1988; Jones *et al*., 1991) suggest that a high percentage of retail-level broiler carcasses are contaminated with *Campylobacter* spp. It is generally assumed that the primary source of initial contamination is the intestinal tract of the chicken. Our findings of 69% prevalence among sampled carcasses are certainly noteworthy. Kapperud *et al.* (1993) examined 176 broiler flocks at slaughter in Norway between April 1990 and April 1991, and found 63% of the flocks were *Campylobacter*-positive during the period from August to November. In a study conducted by Stern and Line (1992), a prevalence of 98% among 50 retail-level chicken carcasses sampled was found. The season of year was not revealed in this study. These findings are comparable with our study, as we found a prevalence of 97% during the month of July. In another related study conducted in the Netherlands, Jacobs-Reitsma *et al.* (1994) reported that *Campylobacter* presence showed seasonal variation, with the highest contamination rate (100%) during the period of June to September, and the lowest (50%) in March. They also indicated that the meteorological data on temperature showed some relation to the presence of *Campylobacter* in broiler flocks; elevated temperatures coincided with high isolation rates. The fact that research findings show a 0 to 100% range of broilers testing positive for *C. jejuni* gives further creditability to a seasonal trend, as is suggested by these studies.

Recently, Stern, (1995) conducted a 2-yr study of broilers obtained quarterly from a processing plant and observed the effect of storage times on seasonal recovery of *C. jejuni*. They found that *C. jejuni* detection decreased after 10 d of refrigerated storage at 4 C. In another published report, Kinde *et al.* (1983) indicated that the presence of *Campylobacter* spp. in market broilers diminishes over time during refrigerated storage. This observation could explain some of this study's results where recovery from the two groups of company broilers obtained and tested on the same date varied. During the month of January, 1 of 15 Company B broilers was positive for *C. jejuni*, whereas 9 of 15 were positive from Company A. The trend was reversed for February, when Company A had the lower positive results (1 of 15), and Company B was higher (13 of 15). Storage time may have reduced the detectability in these broilers. However, there is a significant association found between *Campylobacter* colonization or contamination and farm-dependent factors such as hygiene, different broiler lines, and seasonal flock to flock variability (Stern *et al*., 1990; Willis *et al*., 1991; Jacobs-Reitsma *et al*., 1994). The observations of Stern (1995) of seasonal variation in the levels of *Campylobacter* existence are important in developing intervention strategies. Our findings support the seasonal observation frequencies of *C. jejuni* with broiler carcasses.

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