

Prevalence and Distribution of *Campylobacter* spp. on Poultry and Selected Red Meat Carcasses in Poland

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ABSTRACT

Campylobacter jejuni, *Campylobacter coli*, and *Campylobacter laridis* are recognized causes of alimentary infections in humans. These infections are most often transmitted by foods of animal origin, with undercooked poultry or unpasteurized milk most frequently implicated as vehicles. There are no data on the prevalence and distribution of *Campylobacter* spp. on meats in Poland. We assessed 839 poultry, 105 porcine, and 114 bovine carcasses for the qualitative presence of the organism on the freshly processed product. The organisms were found on 80.3% of the chicken, 48.0% of the duck, 38.0% of the goose, and 3.0% of the turkey carcasses examined. The contamination on porcine and bovine carcasses was 2.9 and 0.9%, respectively. In addition, we assessed and determined that modified *Campylobacter* charcoal differential agar (CCDA) medium was more sensitive and selective than *Campylobacter* brucella agar plate (CAMPY-BAP) medium for the isolation of *Campylobacter* spp. from poultry carcasses.

The primary habitat of *Campylobacter jejuni*, *Campylobacter coli*, and *Campylobacter laridis* is the intestinal tract of warm blooded animals. During slaughter and meat processing these organisms can, and do, contaminate meat products. *C. jejuni* is recognized as a major cause of acute bacteriological gastroenteritis in humans, and consumption of *Campylobacter* spp. — adulterated foods has been associated with many of these illnesses. Studies performed in various countries have shown that between 2 and 14% of the patients manifesting acute gastroenteritis are infected by *C. jejuni* with a frequency comparable to that of *Salmonella* or *Shigella* spp. (3,8,23,25,31,32). *C. coli* and *C. laridis* (nalidixic acid resistant thermophilic *Campylobacter*s — NARTC) are also recognized causes of gastroenteritis, but less frequently than *C. jejuni* (1,14). For these reasons, the presence of these three *Campylobacter* spp. in foodstuffs represents a potential hazard to human health. These three species will be the subject of this paper and will be collectively referred to as *Campylobacter* unless otherwise stated.

Raw or undercooked chickens and other food products of animals origin have repeatedly been implicated in *Campylobacter* induced enteritis (7,12,22). Poultry is a foodstuff with a high incidence of *Campylobacter* contamination (11,20), and as such, represents a potential health hazard for humans. There is limited information available on the prevalence and distribution of *Campylobacter* among ducks, geese, turkeys, pork, and beef carcasses, especially within eastern Europe.

The purpose of this study was to determine the presence of *Campylobacter* on poultry and slaughter animal carcasses in Poland. In addition, we evaluated the effectiveness of two selective media used for isolating *Campylobacter* from meat carcasses.

MATERIALS AND METHODS

Samples

The samples taken consisted of 839 carcasses of poultry (203 chickens, 200 ducks, 200 geese, and 236 turkeys), 105 carcasses of pork, and 114 carcasses of beef. The poultry carcasses were accessible, and were sampled immediately after defeathering and eviscerating, but before chilling in the spin-chiller. We recognized that chilling and further processing could increase or decrease the microbiological quality of the carcasses (34), yet, this was our only access to the carcasses. Pork and beef carcasses were sampled after slaughter, removal of the hide, and dressing of the carcasses, but were taken before chilling.

Direct isolation

Samples for direct isolation were obtained by moistening a sterile cotton swab in Cary-Blair transport medium (15,19) and swabbing a 100 cm² area of the surface. Poultry carcass samples were taken from the following surfaces: breast and the area of the thighs adjacent to the cloacum. Pork and beef carcasses were swabbed on both the ham and shoulder regions. Samples were transported on ice to the laboratory and analyzed immediately upon arrival. Each swab was used to inoculate a *Campylobacter* brucella agar plate (CAMPY-BAP) (3) which was then incubated at 42°C in a microaerobic atmosphere consisting of 5% oxygen, 10% carbon dioxide, and 85% nitrogen for 24 to 48 h. Growth of suspect colonies with typical morphology were screened by examining wet mounts with phase-contrast microscopy. Presumptive positive colonies were streaked for isolation on Brucella-

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FBP [0.025% (w/v) each of ferrous sulfate, sodium metabisulfite, and sodium pyruvate ions] (10) agar. Purified colonies were transferred to semi-solid (0.16% agar) Brucella-FBP medium and incubated at 42°C under microaerobic atmosphere for 24 to 48 h. Each isolate was checked for characteristic morphology, purity, and later identified by taxonomic criteria previously described (15,19).

Whole chicken contamination

Ten consecutive chicken carcasses, taken directly from a mid-morning slaughter line operation after evisceration, were washed by massaging in 250 ml of Cary-Blair transport medium (without agar) in a sterile plastic bag. Next, the rinsings were filtered through a double layer of cheesecloth to remove fat and debris. After filtration, the rinsings were serially diluted in Cary-Blair medium and 0.1 ml was spread-plated onto each of duplicate Campy-BAP plates. The plates were incubated at 42°C under microaerobic conditions for 24 to 48 h. Colonies were counted and estimated for levels of *Campylobacter* contamination on the carcasses.

Comparison of two selective media for direct plating

Fifty-nine carcasses of chickens were swabbed with two swabs, and one swab was used to inoculate each of the two different selective media for direct isolation: Campy-BAP medium (3) and CCDA blood-free selective medium with 32 mg cefobid/L (Pfizer) (11). The plates were incubated under conditions previously described. After incubation, each plate was evaluated for the presence of *Campylobacter* spp. and contamination by other bacterial species.

RESULTS AND DISCUSSION

Isolation rates of *Campylobacter* spp. on poultry, beef, and pork carcasses and enumeration of chicken carcasses are presented in Tables 1 and 2. As shown in Table 1, *Campylobacter* spp. were recovered from 80.3% of the chicken carcasses. Overall levels of contamination ranged from 3.8×10^6 to 3.9×10^7 cfu/carcass. *C. jejuni* was isolated most frequently (54%) and *C. coli* less frequently (39.9%). *C. laridis* was isolated at relatively low frequencies, and was confirmed among only 6.1% of the isolates. Our results indicate that after defeathering and eviscerating the chicken carcass, *Campylobacter* spp. are present in large numbers on the surface of almost all samples taken. These results corroborate data reported by other researchers who isolated *Campylobacter* spp. from 1.8% (27) to 45% (24) to 54 and 62% (20) to 94% (26) of chicken carcasses after slaughter.

Out of 236 turkey carcasses examined, *Campylobacter* spp. were isolated from only 7 (3%) carcasses. The low rate of recovery of these organisms from turkey carcasses differs markedly from results reported by Luechtefeld and Wang (17), who indicated that 33% of edible viscera and 94% of uneviscerated turkey carcasses were positive for *Campylobacter* spp. Moran (18) isolated *C. jejuni* from the internal cavity of 32.3% of turkey carcasses after evisceration, but after spray cleaning the organism was not isolated from carcasses or from any of the remaining sites. The discrepancy in reported results may be due to differ-

TABLE 1. Frequency of isolation of *Campylobacter jejuni*, *Campylobacter coli*, and *Campylobacter laridis* from poultry, pork, and beef carcasses.

Product	No. of isolated strains/ No. sampled (%)	Number/Percent of isolated strains		
		<i>C. jejuni</i>	<i>C. coli</i>	<i>C. laridis</i>
Chickens	163/203 (80.3%)	88/54.0	65/39.9	10/6.1
Ducks	96/200 (48.0%)	61/63.5	17/17.7	18/18.8
Geese	76/200 (38.0%)	40/52.6	32/42.1	4/5.3
Turkeys	7/236 (3.0%)	4/57.1	3/42.9	0/0
Pork	3/105 (2.9%)	1/33.3	2/66.7	0/0
Beef	1/114 (0.9%)	0/0	1/100	0/0

TABLE 2. Recovery of *Campylobacter jejuni*, *Campylobacter coli*, and *Campylobacter laridis* from chicken carcasses by direct plating on Campy-BAP and modified CCDA-Preston media.

Medium	Number of carcasses examined	Incidence—Number/Percent	
		<i>Campylobacter</i> spp.	Contamination by other species
Campy-BAP	59	37/62.7	48/81.4
CCDA	59	55/93.2	5/8.5

ences in colonization rates of the live birds, differences in handling and processing the carcasses, or differences in sample location on the process line. Differences in feeding regimen of the animals could effect shedding or colonization of the organism, and might also account for the differences in isolation rates on the final product.

Results shown in Table 1 indicate that *Campylobacter* spp. were isolated from 48% of duck and 38% of goose carcasses examined. Among the species of *Campylobacter* isolated from ducks, the most frequent was *C. jejuni* (63.5%), followed by *C. laridis* (18.8%), and *C. coli* (17.7%). From geese, the most frequently isolated was *C. jejuni* (52.6%), which was followed by *C. coli* (42.1%), and *C. laridis* (5.3%). Although there have been several studies on the prevalence of *Campylobacter* spp. among chickens and turkeys, there has been limited information on these organisms in ducks and geese (13,16,21). These investigators suggest that ducks may be a potential source of infection for man, as flocks of these birds were colonized at rates ranging from 35 to 100% of those sampled. In those studies carried out by Kasrazadeh and Genigeorgis (13), they reported that 6 to 34% of the duck meat samples yielded *C. jejuni*. They suggest that this comparatively low level of contamination of the duck meat was due to passing the carcass through hot wax, which considerably reduces the number of the organisms on the skin surface and thus decreases the chances of further cross contamination of carcasses.

Previous reports (2,11,16,21,30) have indicated that, for the most part, *Campylobacter* spp. are commensal in the intestinal tract of poultry. Intestinal contents of even

healthy chickens and other species of poultry colonized by *Campylobacter* spp. may contain 10^5 to 10^8 cfu/g. Because such large numbers of *Campylobacter* spp. are released from intestinal contents during the defeathering and eviscerating operations, they are present in large numbers on the poultry carcasses. Scalding times in poultry processing vary from 120 to 170 sec, at temperatures ranging from 58 to 60°C. These time/temperature relationships are inadequate to eliminate all *Campylobacter* spp. contaminating the poultry carcasses, and recontamination occurs again during the defeathering process further serving to contaminate the carcasses. Wempe and co-workers did report that the feather picker and chilling tank were areas of major carcass contamination (34). These are two components of chicken processing which are downstream from the scalding operation and likely account for the levels reported in their study. Further in their study, they reported numbers of the organism exceeding 10^6 /g, indicated that the numbers of *C. jejuni* in the intestinal tract correlated with the presence of the organism in the edible parts.

Our results (Table 1) indicated that our of 105 pork carcasses examined, only 3 (2.9%) yielded *C. coli* (2 of 3) or *C. jejuni* (1 of 3). Higher rates of contamination on pork carcasses or pork meats with *Campylobacter* spp. have been previously reported (6,4,5,28,29). The relatively low rate of contamination reported here is probably due to the prolonged exposure to scalding temperatures of 60°C for 1.5 to 4 min. These conditions appear to be relatively stringent, and would account for killing *Campylobacter* spp. on the skin of the carcass.

As with the case with porcine carcasses, contamination of bovine carcasses with *Campylobacter* spp. was low. Of 114 carcasses examined, only 1 *C. coli* was isolated (Table 1). These results agree with those reported by others (9,28,33) who found low levels of the samples examined to yield *Campylobacter* spp.

Table 2 illustrates the results from the experiment comparing Campy-BAP (3) and CCDA (11) media for isolating *Campylobacter* spp. from chicken carcasses. These results show that by use of CCDA medium we recovered *Campylobacter* spp. from 93.2% of the carcasses examined, while by using Campy-BAP medium we recovered the organism from only 62.7% of the samples. The rates for the non-*Campylobacter* spp. flora on CCDA medium was 8.5% of the colonies picked, while on Campy-BAP the breakthrough flora accounted for 81.4% of the colonies picked. These results indicate that modified CCDA medium is more sensitive and selective than Campy-BAP for isolation of *Campylobacter* spp. from chicken carcasses.

In conclusion, our data provide evidence showing that poultry carcasses appear to be prominent reservoirs of *Campylobacter* spp. in Poland. Both pork and beef carcasses are less frequently contaminated with the organism. The next issue to be resolved is the epidemiologic link between consumption of these food products and human disease. As Poland is a country with different husbandry and production procedures if compared to western Europe and the United States, the transmission of the organism to humans

via these food products cannot be assumed. Rather, serotyping data comparing isolates of *Campylobacter* spp. from human disease with those isolated from poultry would be needed. In the United States, large outbreaks of this disease are rare. Rather, the disease is most frequently manifested in sporadic, individual cases. Therefore, outbreak epidemiology in the classic manner of investigation would seem an inappropriate method to understanding the transmission of the organism to humans in Poland. If serotypes of *Campylobacter* spp. from humans and from poultry are similar, more confidence would be ascribed attributing this vehicle to disease transmission in Poland. Decontamination of the poultry carcasses, or diminishing the presence of the organism in live birds would then be required to assist in protecting the public health.

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