Intestinal Carriage of *Campylobacter jejuni* and *Salmonella* by Chicken Flocks at Slaughter

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ABSTRACT

***Campylobacter jejuni* were isolated in large numbers from the majority of birds sampled in colonic swabs from 28 of 60 flocks at slaughter. By contrast only small numbers of birds from 11 of the same 60 flocks yielded *Salmonella enteritidis* serotypes. Three *C. jejuni* isolates from each flock were serotyped on the basis of their heat-stable antigens, using antisera prepared against 16 serotypes common in *Campylobacter* diarrhea in man. The majority (72 of 83) of the chicken isolates could be serotyped.**

Key words: *Campylobacter jejuni* serotypes, *Salmonella enteritidis*, chickens.

RÉSUMÉ

Cette expérience impliquait 60 troupeaux de poulets de gril et elle consistait à prélever deux écouvillons du côlon de dix sujets de chacun d'eux, lors de l'abattage. Les auteurs réussirent ainsi à isoler une flore luxuriante de *Campylobacter jejuni*, chez la majorité des dix poulets de 28 des 60 troupeaux. Par ailleurs, seulement quelques sujets de 11 des 60 troupeaux précités recelaient des sérotypes de *Salmonella enteritidis*. Les auteurs tentèrent aussi de déterminer le sérotype de trois souches de *C. jejuni* de chacun des troupeaux, en se basant sur leurs antigènes thermorésistants et en utilisant des antisérums contre 16 sérotypes communs de *C. jejuni* responsables de diarrhée, chez les humains. Ils réussirent ainsi à déterminer le sérotype de 72 des 83 souches qu'ils avaient isolées.

Mots clés: sérotypes de *Campylobacter jejuni, Salmonella enteritidis*, poulets.

Both *Campylobacter jejuni* and *Salmonella enteritidis* serotypes are important causes of gastrointestinal disease in man (1). Much is known about the epidemiology of salmonellosis in chickens and its transmission to man (2), but similar knowledge about *Campylobacter* in chickens and its relation to human illness is relatively slight. The recent development of serotyping systems for *C. jejuni* (3,4) gives the opportunity to determine whether chickens are likely to be an important reservoir of infection for man. The purpose of the present study was to compare the prevalence of *C. jejuni* to that of *Salmonella* in Ontario chicken flocks at slaughter and to determine the serotypes of *C. jejuni* involved using a serotyping system based on determination of heat-stable antigens.

Two swabs were taken from the colon of each of ten chickens from 60 different flocks at slaughter. Swabs were stored in Cary-Blair transport medium at 4°C for up to 48 hours before culture in the laboratory. One swab was streaked and cultured microaerophilically on the selective medium Campy-BAP for 48 hours for *C. jejuni*, which were identified by methods previously described (5). The other swab was used for *Salmonella* isolation. Swabs were inoculated into 5 mL of peptone water and incubated for 24 hours at 37°C; one mL of the peptone water was inoculated into 9 mL of tetrathionate broth, which was incubated for 24 hours at 42°C before a loopful of the broth was subcultured onto brilliant green agar (6). *Salmonella* were identified by specific phage lysis and the isolates serotyped (7).

The heat-stable antigens of *C. jejuni* isolated were identified by titration using antisera prepared against the serotypes commonly found in diarrheic illness in man, i.e. 1, 2, 3, 4, 5, 8, 10, 11, 13, 16, 18, 19, 21, 23, 31 and 37 (3,5). Only three *C. jejuni* isolates, randomly chosen from each group of ten birds, were serotyped.

Salmonella enteritidis isolations — Thirteen of 110 birds from 11 of the 60 flocks yielded *Salmonella* by enrichment culture of colonic swabs. The serotypes isolated were *S. infantis* (three flocks), *S. mbandaka* (three), *S. heidelberg* (two), *S. meleagridis, S. schwarzengrund* and *S. nienstedten* (one flock each). In only two groups were two birds culture-positive for *Salmonella*; the other nine groups had only one bird positive.

Campylobacter jejuni isolations — Two hundred and fifty-eight of 280 birds from 28 of the 60 flocks were carriers of *C. jejuni*. The other 32 flocks yielded no *C. jejuni*. The numbers of birds, in each group of ten, yielding *C. jejuni* were ten (21 flocks), nine (three flocks), eight (one), four (one) and two birds (one). Four groups yielding *Salmonella* also carried *Campylobacter*.

The results of serotyping three isolates from each group are given in Table I. In 11 groups three of three isolates were the same serotype, in 12 two of three were the same, and in five groups the isolates were of different serotype. From only one group were all three isolates untypable with the antisera used, in two others two were untypable, and in four others one was untypable. Within some groups of

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TABLE I. Numbers of Flocks Carrying Different Serotypes of *Campylobacter jejuni*, and Number of Isolates of Each Serotype Made in 28 Flocks

Description	Serotype										
	1	2	3	4	8	11	13	21	37	NT[a]	Other
No. of flocks with each serotype (total 28 flocks)	9	3	4	2	4	4	5	2	2	7	8[b]
No. isolates made (total 83 isolates)	18	5	7	2	6	4	9	4	5	11	12[b]

[a]Nontypable
[b]2, 8 (2 flocks, 1 isolate): 1, 8 (3); 8, 3 (2); 5, 21 (2); 5, 16 (1); 23, 1 (1); 11, 5, 8 (1). (In parentheses, number of isolates from each group of 3 birds tested)

TABLE II. Other Antigens Inconsistently Identified in Some Flocks Together with the Major Antigens (Serotype)

Major or minor antigen	Flock number					
	32	35	36	45	58	59
Major antigen (number of isolates)	11 (1)	2 (2)	2 (2)	8 (1)	5,21 (2)	1 (2)
Other antigens identified (number of isolates)	11,5,8 (1)	2,8 (1)	2,8 (1)	8,3 (2)	5,16 (1)	1,23 (1)

birds extra antigens were inconsistently detected in some of the *Campylobacter* isolates, either as major or minor antigens. A minor antigen is defined as one giving a titer of more than two dilutions below that of the homologous serotype used to raise the antiserum. These other antigens are shown in Table II.

There is epidemiological evidence that the chicken is an important reservoir of *Campylobacter* infection for man (8-12). Our study has shown that nearly half of 60 chicken flocks cultured had large numbers of *C. jejuni* in their intestinal contents. This high carriage was in marked contrast to that of *Salmonella*, where only individual birds in relatively few flocks carried the organism. Our findings are similar to a recent Hungarian study (13).

The chicken slaughtering and cleaning process is associated with contamination of carcasses with intestinal bacteria (14), so that many carcasses on retail sale are contaminated with *Campylobacter* (reviewed elsewhere, 1). Our study suggests that *Campylobacter* are likely to be found in larger numbers on chicken carcasses than *Salmonella*. Nevertheless, only half the flocks tested carried *C. jejuni* and a previous study in Ontario found only a quarter of flocks to be infected (5). This suggests that hygienic husbandry practices could be used to control *Campylobacter* infection in chickens.

In two Canadian studies the most common heat-stable serotypes of *C. jejuni* involved in human diarrheic illness were, 2, 4, 3, 1, 8 and 13/16 (61% of isolates) (15) and 4, 2, 1, 5, 8 and 13/16 (51% of isolates) (16); other frequent but less common isolates serotypes were 18, 31 and 45 (16) and 5, 18, 21 and 36 (15). We identified eight of these serotypes (sometimes with other antigens) in the majority of the flocks. We assume that serotype 13 was in fact 13/16 but that our relatively low titered antisera against serotype 16 was inadequate to detect the antigen. In only seven flocks were untypable isolates encountered.

A previous study of isolates from seven chicken flocks identified serotypes 1, 2, 3, 4, 5 and 31 (5). In the previous study four of seven flocks had only one serotype present among ten isolates. For this reason we only serotyped three of the isolates from each flock, but more than one serotype was observed in 17 of the 28 flocks. It is thus possible that *Campylobacter* infection in man acquired from chicken could be associated with more than one serotype. Outbreaks of *C. jejuni* infection in man involving more than one serotype have been observed and individuals involved have shown serological response to more than one of the serotypes (17).

Campylobacter jejuni has thus been found in large numbers in nearly half of Ontario chicken flocks at slaughter, where it was present more frequently and in larger numbers than *Salmonella*. The serotypes (heat-stable antigens) commonly recovered were those often found in human enteric disease suggesting that the chicken can be an important source of infection for man.

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