Distribution of serotypes of Campylobacter jejuni and C. coli from Danish patients, poultry, cattle and swine

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Abstract

The number of human cases of enteritis caused by Campylobacter jejuni and C. coli is increasing in Denmark and other European countries. No systematic typing has earlier been performed on Campylobacter isolates of Danish origin. The primary purpose of this study was to provide a serotype distribution of Campylobacter isolates from Danish patients and the major food production animals. In addition, the occurrence of intestinal carriers of thermophilic campylobacters among these food production animals was examined. In a nationwide survey, the individual isolation rate was 36% for broiler chickens, 47% for cattle and 46% for swine when sampled at the slaughterhouse. C. jejuni accounted for 83–91% of the thermophilic Campylobacter spp. in broiler chickens and cattle, whereas 95% of the isolates from swine was C. coli. In human patients with Campylobacter enteritis, 94% of the isolates were C. jejuni and 6% were C. coli. Heat-stable serotyping (the 'Penner scheme') was performed on a total of 398 isolates from the four sources: human patients (n = 145), broiler chickens (n = 94), swine (n = 111) and cattle (n = 48). Among human isolates, serotype O:1,44, O:2 and the O:4-complex accounted for 62% of the C. jejuni isolates. These serotypes were also common in samples from broilers and cattle. In swine, C. coli O:30 and O:46 were most common. The serotype distribution of human clinical isolates showed large overlap with the serotype distribution of campylobacters in cattle and chickens, and on this basis both could be major sources of human campylobacteriosis.

Keywords: Campylobacter jejuni; Campylobacter coli; Serotyping; Prevalence

1. Introduction

In many developed countries, Campylobacter is the most common or the second most common bacterial enteric pathogen [1,2]. In Denmark, thermophilic Campylobacter spp. account for an increasing number of the cases of bacterial enteritis as the annual number of registered cases of campylobacteriosis has doubled during the last four years: from 1100–1500 cases in 1982–1992 to 2973 cases in 1996 [3]. The frequency of the most common zoonotic infection in Denmark, salmonellosis, showed a very different development during the same years with stagnation or a small decrease in 1994–1996, and the increase in campylobacteriosis cannot be related to the number of samples analyzed or the methods of analysis. The major sources of Campylobacter infections in the
United States and other developed countries are assumed to be poultry, raw milk, untreated surface water and pets [2]. However, except for large outbreaks, the actual source of infection is rarely identified. Nevertheless, undercooked poultry is assumed to be the single most important cause of sporadic cases, but other foods are also very likely sources as campylobacter can be isolated from many different types of foods, for example raw beef, pork, lamb, cooked meats and seafood [4, 5].

Serotyping or other typing methods are necessary epidemiological tools in studies of zoonotic bacteria. For example, combined serotyping and phage-typing of salmonella isolates from humans, layer hens, broiler chickens, swine and cattle were used for assessing the significance of different production animals as a source of human infection [6, 7]. For campylobacters, comparison of types isolated from humans and potential sources of human infection has not been done over long time periods and to the same extent. In the 1980s Campylobacter serotypes from chickens, cattle and swine were compared with the most common serotypes from human patients in Canada [8], serotypes of campylobacters from patients, swine and cattle were compared in Holland [9, 10], and in England serotypes of campylobacters from food and environmental samples were compared with isolates from patients [4].

Two serotyping schemes for thermophilic campylobacters have been established, i.e., the ‘Lior scheme’ based on heat-labile (HL) antigens (surface proteins) [11] and the ‘Penner scheme’ based on heat-stable (HS) antigens [12]. It was later shown that lipo-polysaccharide (LPS) conferred the sero-specificity of the Penner serotyping system similar to the O-serotypes of Enterobacteriaceae [13–15]. However, the HS-antigens of Campylobacter were shown to be of an unusual structure and chemical composition. Some C. jejuni and all C. coli serostrains and wild-type strains examined so far produce the usual high molecular mass LPS-structure composed of lipid A, core oligosaccharides and an O side-chain, i.e., a polymer of repeating oligosaccharide units [16, 17]. But more than half of the C. jejuni serotype reference strains (serostrains) only produce low molecular mass LPS that lacks an O side-chain [17]. This structure resembles the lipo-oligos saccharides (LOS) of a few other Gram-negative bacteria such as Neisseria and Haemophilus spp. [18]. The serotype differences (HS-antigens) in strains possessing the LOS-structure are reflected in variations in the outer core attached to an inner region of invariable structure [19]. In contradiction to these studies, it has recently been suggested that the HS-antigens of the Penner serotyping system are capsular [20].

Other phenotypic typing methods than serotyping, e.g., biotyping and phage-typing, are developed for campylobacters and have been used in epidemiological studies [21–23]. Many different genotypic typing methods, e.g., restriction endonuclease analysis, ribotyping, pulsed field gel electrophoresis and PCR-fingerprinting, have been used for typing of campylobacter and have proven useful for human outbreaks of campylobacteriosis [24–27] and epidemiological studies in for example broiler flocks [28] and swine farms [29]. The problem with the genotypic typing methods is that they are rarely standardized and types can therefore not be compared between laboratories. Although antisera for the two serotyping systems are not commercially available, the same reference strains are used for production of antisera, and the serotypes should be comparable between different laboratories.

As there are no previous reports on the serotype distribution of Danish campylobacters, the purpose of the present study was to provide such a serotype distribution in campylobacters isolated from human patients and from food production animals that are

<table>
<thead>
<tr>
<th>Table 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of fecal samples from cattle, broiler chickens and swine examined for campylobacters. Number and percentage of samples positive for thermophilic campylobacters and the distribution of species.</td>
</tr>
<tr>
<td>No. of slaughterhouses (samples from each)</td>
</tr>
<tr>
<td>---</td>
</tr>
<tr>
<td>Cattle</td>
</tr>
<tr>
<td>Chicken</td>
</tr>
<tr>
<td>Swine</td>
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</tbody>
</table>
potential sources of human infections. This study is part of a continuous national survey that examines the occurrence of zoonotic bacteria in poultry, swine and cattle. For Campylobacter, the species and heat-stable serotype ('Penner serotype') distribution were also determined. Results of the first year of the campylobacter survey are reported here. Species and serotype of Campylobacter isolated from human patients with diarrhoea in the same period are included for comparison.

2. Materials and methods

2.1. Sampling

As part of a continuous survey of zoonotic bacteria in Danish cattle, poultry and swine, samples were collected at the slaughterhouses in the last quarter of 1995 and the first three quarters of 1996. Faecal samples from cattle and caecal contents from swine were sampled four times during the year at each slaughterhouse, whereas cloacal swabs from broiler chickens were collected each week. Sampling was designed to obtain a representative number of samples from all parts of Denmark: slaughterhouses were distributed over the entire country, the number of samples taken at each slaughterhouse varied according to its number of slaughters, and only one sample was taken from each flock of chickens and each herd of cattle or swine. The number of samples and slaughterhouses are shown in Table 1.

Thermophilic Campylobacter strains isolated from human patients with diarrhea were randomly selected from Statens Serum Institut’s routine samples from the second half of 1995 and the first half of 1996. Only one isolate from each patient and one isolate from a known outbreak was included.

2.2. Isolation and identification

Samples from cattle and swine were sent to the laboratory by mail (usually 1 day in the postal system). Upon receipt at the laboratory, the samples were stored at 4°C until culturing. Most samples were cultured 1–3 days after they were taken at the slaughterhouse, but in some cases up to 5 days after

| Table 2 |
| Distribution of serotypes among C. jejuni isolates from humans, chickens and cattle |

<table>
<thead>
<tr>
<th>Serotype</th>
<th>Human (n = 136)</th>
<th>Chicken (n = 75)</th>
<th>Cattle (n = 45)</th>
</tr>
</thead>
<tbody>
<tr>
<td>O1,44</td>
<td>25% 18%</td>
<td>11% 15%</td>
<td>4% 9%</td>
</tr>
<tr>
<td>2</td>
<td>36% 26%</td>
<td>20% 27%</td>
<td>14% 31%</td>
</tr>
<tr>
<td>3</td>
<td>6% 4%</td>
<td>1% 1%</td>
<td>–</td>
</tr>
<tr>
<td>4-complex</td>
<td>24% 18%</td>
<td>10% 13%</td>
<td>4% 9%</td>
</tr>
<tr>
<td>5</td>
<td>5% 4%</td>
<td>2% 3%</td>
<td>2% 4%</td>
</tr>
<tr>
<td>6,7</td>
<td>2% 1%</td>
<td>4% 5%</td>
<td>3% 7%</td>
</tr>
<tr>
<td>8</td>
<td>2% 1%</td>
<td>–</td>
<td>1% 2%</td>
</tr>
<tr>
<td>11</td>
<td>3% 2%</td>
<td>2% 3%</td>
<td>4% 9%</td>
</tr>
<tr>
<td>12</td>
<td>3% 2%</td>
<td>1% 1%</td>
<td>–</td>
</tr>
<tr>
<td>19</td>
<td>3% 2%</td>
<td>3% 4%</td>
<td>3% 7%</td>
</tr>
<tr>
<td>21</td>
<td>1% 1%</td>
<td>2% 3%</td>
<td>3% 7%</td>
</tr>
<tr>
<td>23,36</td>
<td>2% 1%</td>
<td>3% 4%</td>
<td>3% 7%</td>
</tr>
<tr>
<td>29</td>
<td>1% 1%</td>
<td>–</td>
<td>2% 2%</td>
</tr>
<tr>
<td>37</td>
<td>4% 3%</td>
<td>1% 1%</td>
<td>–</td>
</tr>
<tr>
<td>42</td>
<td>2% 1%</td>
<td>–</td>
<td>1% 2%</td>
</tr>
<tr>
<td>53</td>
<td>4% 3%</td>
<td>–</td>
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</tr>
<tr>
<td>55</td>
<td>4% 3%</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Others***</td>
<td>3% 2%</td>
<td>7% 9%</td>
<td>–</td>
</tr>
<tr>
<td>Non-typable</td>
<td>6% 4%</td>
<td>8% 11%</td>
<td>1% 4%</td>
</tr>
</tbody>
</table>

Results of C. jejuni isolated from swine: four C. jejuni isolates were serotyped: O:2, O:53, O:23,36, O:23,36,5.

* Cattle: in five cases, more than one strain was isolated from the same sample. As these had different serotypes, they are included in this table.

** An isolate is ascribed to the O:4-complex if it reacts in one or more of the sera: 4, 13, 16, 43, 50, 64.

sampling. Cloacal swabs from broiler chickens were collected from live birds prior to stunning, placed in transport medium [30], and sent to the laboratory. Culturing was initiated the same day as received at the laboratory (usually 24 h after sampling, but in some cases up to 3 days after sampling).

The presence of thermophilic *Campylobacter* spp. in broiler chickens was determined by streaking the cotton swab onto modified charcoal cefoperazone deoxycholate agar (CCDA) (Campylobacter Blood-Free Selective Agar Base (Oxoid, Basingstoke, UK) with CCDA Selective Supplement (Oxoid, SR155E)). Swine and cattle samples were cultured both by selective enrichment and direct plating. For selective enrichment, 1 g of the sample was added to 10 ml Preston broth: Nutrient Broth no. 2 (Oxoid) supplemented with 5% lyed horse blood, Campylobacter Growth Supplement (Oxoid, SR84E) and Preston Campylobacter Selective Supplement (Oxoid SR117E). After incubation under microaerobic conditions (approx. 6% O2, 7% CO2, 7% H2, 80% N2) for 18–24 h at 42°C, one loopfull (10 μl) of the broth was transferred to CCDA. The plates were incubated microaerobically at 42°C and inspected after 1–2 days. Negative plates were reincubated for up to four days. Clinical isolates from human patients were isolated on modified Skirrow agar (Statens Serum Institut, Denmark), incubated microaerobically at 37°C for 48 h.

One colony from each sample was subcultured and identified to the species level by the use of phase-contrast microscopy (characteristic morphology and mobility), catalase, oxidase, indoxyl acetate hydrolysis, hippurate hydrolysis, susceptibility to nalidixic acid and cephalothin [31]. Samples with *C. coli*, *C. jejuni* or *C. lari* were recorded as positive for thermophilic campylobacter.

2.3. Production of antisera

Reference strains for the Penner serotyping scheme for HIB-antigens were obtained from the Culture Collection of the University of Göteborg (CCUG), Sweden. Antisera were prepared according to a slightly modified version of the method described by Jacobs-Reitsma et al. [32]. Briefly, New Zealand white rabbits (3–3.5 kg; Hvidesten, Denmark) were immunized intravenously four times (on day 1, 6, 11 and 15 with 0.25 ml, 0.5 ml, 1.0 ml and 2.0 ml, respectively) with a suspension of the reference strain with an OD650 nm of 0.35–0.40 (approx. 5×10^8 cfu/ml). The bacteria were treated with formalin for 2 h and 24 h, respectively, before the first and second immunization. Live cells were used for the last two immunizations. The rabbits were bled six days after the last immunization. Antisera against the following 49 serostrains were prepared: 1–13, 15–19, 21–27, 29–31, 34, 36–46, 48, 50, 53–57, 59, 64. According to the serotyping scheme with separate sets of anti-

<table>
<thead>
<tr>
<th>Serotype</th>
<th>Human <em>(n = 9)</em></th>
<th>Chicken <em>(n = 19)</em></th>
<th>Swine <em>(n = 107)</em></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><em>n</em></td>
<td>%</td>
<td><em>n</em></td>
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<tr>
<td>5</td>
<td></td>
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<td>4</td>
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<td>24</td>
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<td>11</td>
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<td>25</td>
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<tr>
<td>46</td>
<td>1</td>
<td>11</td>
<td>1</td>
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<tr>
<td>48</td>
<td>1</td>
<td>11</td>
<td>1</td>
</tr>
<tr>
<td>54</td>
<td>1</td>
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<tr>
<td>56</td>
<td>1</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>59</td>
<td>1</td>
<td>11</td>
<td></td>
</tr>
<tr>
<td>Non-typable</td>
<td>0</td>
<td>0</td>
<td>4</td>
</tr>
</tbody>
</table>

Results of *C. coli* isolated from cattle: three *C. coli* isolates were serotyped: two O:48 and one non-typable.
sera for *C. jejuni* and *C. coli* [33], this gives 38 *C. jejuni* serotypes and 12 *C. coli* serotypes (as O:5 is common to *C. jejuni* and *C. coli*). The total system consists of 47 *C. jejuni* serostrains and 19 *C. coli* serostrains. The antisera were chosen on the basis of the most prevalent serotypes among human patients and food animals reported in Europe and North America [32,34–36].

2.4. Serotyping

*C. jejuni* and *C. coli* isolates were serotyped by passive haemagglutination in microtiter plates as described by Penner and Hennessy [12], but only three dilutions of antisera were used (1 in 80, 1 in 640, 1 in 5120). *C. coli* and *C. jejuni* were first serotyped with separate sets of antisera, but if a strain had no reaction with these it was tested against all antisera. All cattle strains isolated during one year were serotyped (48 strains) and a random selection of 111 swine isolates, 94 chicken isolates and 145 human isolates were serotyped.

2.5. Statistical methods

$\chi^2$ tests were performed to test for differences in frequency of serotypes between groups. SAS statistical software (version 6.11; SAS Institute, Cary, NC, USA) was used.

3. Results

3.1. Isolation rates and distribution of species in cattle, chickens and swine

The survey showed that 47% of cattle, 46% of swine and 36% of broiler chickens were positive for thermophilic campylobacters, i.e., *C. jejuni*, *C. coli* or *C. lari*. Mostly *C. coli* was found in swine, whereas *C. jejuni* was the most abundant species in cattle and broilers (Table 1). Very few *C. lari* were isolated from all sources. Twenty-four percent of cattle samples and 5% of swine samples were positive for *Campylobacter hyointestinalis* (data not shown). This finding was unexpected as the selective culture conditions of CCDA (32 mg l$^{-1}$ of cefoperazone) and incubation at 42°C generally does not support growth of *C. hyointestinalis*. As the colonies of *C. hyointestinalis* clearly had a slower growth, i.e., smaller colonies, on CCDA, it was fairly easy to select a colony of a thermophilic species if these were present. Therefore, as only one colony was selected from each sample, *C. hyointestinalis* was primarily isolated from samples that were negative for thermophilic *Campylobacter* spp.

Of the 145 randomly selected human isolates, 136 (94%) were *C. jejuni* and 9 (6%) were *C. coli*.

3.2. Serotypes of *C. jejuni* and *C. coli*

Most strains reacted in only one serum or in combinations of sera comprising well-known complexes, e.g., O:1,44; O:4,13,16,43,50,64 (in the following designated the O:4-complex), O:6,7 and O:23,36. To simplify tables, a strain reacting in one or more of the sera of one of these complexes was ascribed to that serotype complex. This is justified by studies showing that isolates belonging to the same clone can vary in their reactions in the antisera of for example the O:4-complex [37]. One fourth of the *C. coli* from swine and a few *C. jejuni* reacted in two or more antisera which were not in well-known complexes. For these, all major reactions were recorded (i.e., a weak reaction at the lowest serum dilution (1 in 80) was not included), but the strain was classified as belonging to the serotype of the highest titre. Of course, this grouping reduces the number of different serotypes, but for the present purpose of obtaining an overview of the serotype distribution among isolates from different sources this simplification is advantageous. However, in outbreak situations it would be important to compare the exact reactions.

The three most common *C. jejuni* serotypes in human patients were O:2, O:4-complex and O:1,44, accounting for 62% of the isolates, whereas the other serotypes each represented 4% or less (Table 2). In broilers, the same three serotypes were dominant, accounting for 50% of the isolates. In cattle, O:2 was the dominant serotype (31%) with seven other serotypes being relatively common, representing 7–9% each (O:1,44, O:4-complex, O:6,7, O:11, O:19, O:21, O:23,36). The differences in serotype distribution between strains of human, chicken and cattle origin were not statistically significant. In all sources,
a wide variety of serotypes were found: in humans 18 different serotypes or serotype complexes, in chickens 15 and in cattle 12 (not including different combinations of reactions in for example the O:4 complex). Of the 38 C. jejuni antisera used, six sera were not found to react with any of the isolates in this study (9, 38, 40, 41, 45 and 57).

Fig. 1 shows the eight most common human C. jejuni serotypes and the representation of these in cattle and chickens. These serotypes cover 79% of the human isolates, but 56% and 53% of chicken and cattle isolates, respectively. It should be noted that 14 antisera are needed to cover all reactions included in these serotypes and serotype complexes.

Only nine human C. coli strains were isolated, and these had seven different serotypes (Table 3). The most common C. coli serotypes from chickens were O:30 (26%) and O:5 (21%). In swine, O:30 and O:46 were most common, but four other serotypes represented more than 10%: O:5, O:24, O:54, O:56 (Table 3). Especially the swine C. coli isolates tended to have complex serotypes, i.e., one isolate had strong reactions with two or three antisera. Four isolates had serotype O:24,54, but otherwise each complex only represented one or two isolates.

The relationship between the age of the patient and the serotype isolated is shown in Fig. 2. Patients were divided into four age groups: 0–4 years (n = 31), 5–19 years (n = 19), 20–29 years (n = 43) and ≥30 years (n = 52). Only the three most common serotypes were included in this analysis: O:1,44; O:2 and O:4-complex, each constituting 18–26% of all isolates from patients. Serotype O:1,44 was not found in age group 5–19 years, while this serotype constituted 16–26% in the other age groups. This difference was statistically significant (P = 0.03). In contrast, age group 5–19 years had relatively more strains of serotype 2 (42%). It should be noted that there is a relatively low number of patients in age group 5–19 years. There was a tendency to increased frequency of the O:4-complex with increasing age: from 6% among 0–4 year-olds to 21% in patients 30 years or older (Fig. 2). These differences were not statistically significant as tested by the χ² test (P = 0.06–0.08).

3.3. Typability

With the use of three-fourths of the C. jejuni/coli antisera in the HS-serotyping scheme, the typability was 95%, 81%, 94% and 99% for strains of human, chicken, cattle and swine origin, respectively. The number of non-typable strains was significantly different (P = 0.001) between sources. The antisera were selected primarily on the basis of serotyping results of human strains in other countries and to a lesser extent from animal sources. It is therefore not surprising that a good typability was obtained for the human strains, whereas the chicken strains had a lower typability with these selected sera.

4. Discussion

Intestinal carriers of thermophilic campylobacters were very common among Danish broiler chickens, cattle and swine. About half of all samples were found positive for campylobacters. For chickens, this level of carriers was expected. In a Canadian survey, thermophilic campylobacter was found in 38% of carcasses of broiler chickens sampled from the final chill tank at the slaughterhouse [38]. In Dutch breeder farms, campylobacters were isolated in 67% of flocks when 30 animals were sampled from each flock, and the prevalence within positive flocks ranged from 20 to 100% [39]. In the present study, only one animal from each flock was sampled. In a one-year study in 1987–1988 in Sweden, 27% of 287 broiler flocks at 18 farms were campylobacter positive (40 samples per flock) [40]. In a follow-up of this Swedish study, the prevalence decreased to 16–10% in 1988–1990 [41]. The decrease was assumed to be due to increased awareness of hygiene routines among the Swedish farmers participating in the study.

We found that 46% of swine had detectable levels of thermophilic campylobacter, mostly C. coli, in the caecal content at slaughter. This is a relatively low isolation rate compared to other studies. Weijtens et al. [29] found that 85% of swine from eight Dutch farms were carriers of thermophilic Campylobacter when ileal and rectal samples were collected at the slaughterhouse. Another Dutch study found a rate of 79% of intestinal carriers (caecal samples) at three slaughterhouses [42]. In the UK, the isolation rate of campylobacters was 47% in healthy swine and 77% in faeces of swine with symptoms of enteric
disease [43]. In at least one of the Dutch surveys, culturing was initiated immediately (less than 3 h) after sampling [29], whereas samples of the present study and the UK study were sent through the post at ambient temperature without transport medium. This delay is likely to decrease the number of culture positive samples as the average level of campylobacters in faeces of swine right after slaughter is only around 10^3 cfu g^-1 [29], and the survival of campylobacters is poor at temperatures between 20 and 30°C [44,45] and even at 4°C for 24 h the number of culture positive faecal samples is reduced significantly [46]. It should also be noted that just one sample was taken from each stock of swine/cattle, whereas the other studies mentioned have multiple sampling from each herd. Thus in these, the isolation rate is both a measure of the frequency of campylobacter positive animals in each herd and the prevalence among herds. As this study is part of a continuous survey of Campylobacter and other less sensitive bacteria in geographical widespread slaughterhouses, it was not possible to obtain better conditions for isolation of campylobacters.

In cattle, the isolation rate of thermophilic campylobacter, primarily C. jejuni, was 47%. In addition, C. hyointestinalis was found in 24% of the samples. This is in good agreement with the results reported by Grau [47], who found C. jejuni and C. hyointestinalis in the feces of 54% and 88%, respectively, of young calves (about 4 weeks of age) and 13% and 47%, respectively, of adult cattle in Australia. Contrary to the present study, the culture techniques of the Australian study were aiming at isolating both C. hyointestinalis and C. jejuni. In Canada, Garcia et al. [48] found a C. jejuni isolation rate of 39% in large intestinal contents from steers at slaughter.

The prevalence of intestinal carriers among food production animals is not directly reflected in the prevalence of Campylobacter in food products at the retail level. In Denmark, 25-44% of poultry products were contaminated with thermophilic Campylobacter spp., but only 0.6–2% of beef and pork were contaminated [49]. A UK study also showed highest prevalence of campylobacters in poultry (56%), but also relatively high prevalence in beef (24%), pork (18%), lamb (16%) and seafood (15%) [4]. It has been shown that the number of campylobacters on swine carcasses is significantly reduced during cooling with forced ventilation, resulting in no detectable campylobacter on the carcasses after storage overnight [50]. This reduction is most likely due to drying of the skin surface.

Ninety-five percent of the human strains and 81–99% of the strains of animal origin could be serotyped with 49 of the 65 antisera in the HS-serotyping system. A high typability is usually found for human strains: in the USA 98% were typable with 57 antisera [51] and 85% were typable with the 24 most common antisera [35], in Sweden 80% of human isolates could be typed with only 22 sera [36]. The overall typability of campylobacter sampled in Dutch epidemiological studies of poultry was 81% with the use of all 65 absorbed antisera [32], which is the same typability as in the present study. It should be noted that even when strains are tested against all of the 65 antisera, some strains are untypable, indicating that the serotyping scheme is not yet complete.

The most common serotypes of human origin were O:2, O:1,44 and the O:4-complex. This is in good agreement with the serotypes found in Swedish patients in 1984–1985, where these serotypes accounted for 33%, 9% and 9%, respectively, of 45 isolates obtained from patients who were assumed infected in Sweden [36]. The serotype distribution in Swedish patients infected abroad was slightly different. In northern Norway, serotyping of 79 human isolates in 1980–1983 showed that the O:4-complex, O:6,7 and O:12,40 were the most common serotypes, whereas only one isolate of O:2 was typed [34]. Contrary to the Norwegian study, no O:12,40 and only two O:12 were found among the human isolates in the present study. In general, the most common serotypes among these Danish isolates from patients were also common serotypes in Britain [52], USA [35], Canada and other countries worldwide (J.L. Penner, personal communication). However, the ranking was not the same, e.g., serotype O:8 ranked third in the British and the USA study, whereas only two isolates of this type were found among the Danish isolates.

It has not been possible to find clear relationships between HS- or HL-serotypes and clinical symptoms among human patients [53], and generally little is known about the role of different surface structures in the pathogenicity of campylobacters. However, we
found a weak relationship between the age of the patient and the serotype of the isolate, which could indicate presence of serotype related differences in the ability to cause disease in different hosts. Isolates from a larger number of patients would be necessary to study this possible relationship in more detail. Among children of the age 0–10 years, Lastovica et al. [54] also found differences between HS-serotypes and age group of the children.

In Holland, epidemiological studies in poultry showed that O:2, O:37, O:1,44 were the most common ‘C. jejuni serotypes’ and O:59, O:56, O:30 were the most common ‘C. coli serotypes’ [32]. The strains were not speciated in this study, and all thermophilic isolates were serotyped using the complete set of 65 absorbed antisera. An early study in Canada showed that O:1, O:2, O:3, O:5 and O:31 were the most frequent serotypes among C. jejuni isolates from chickens [8]. Only the first two of these were also common in the present study.

Fricker and Park [4] found that the serotypes most common in human patients were common serotypes in a number of different food samples: beef, poultry, lamb, offal and seafood, whereas other serotypes were found in pork. As in the present study, Dutch and Canadian studies found that the most frequently occurring serotypes in human patients were also common in poultry and cattle, but not in pigs [8–10]. In the present study, the species distribution of human clinical isolates, 94% C. jejuni and 6% C. coli, indicates that both poultry and cattle could be major sources of the human Campylobacter enteritis cases, whereas swine are not likely to be one of the most important sources, as only 4% of the swine isolates were C. jejuni. The serotyping results showed a large overlap of the most common serotypes in humans, cattle and broiler chickens. Thus, food products originating from cattle and chickens could both be important sources of human infections. The high prevalence of campylobacters in poultry products at the retail level in Denmark [49] makes these products a likely candidate as one of the major sources for zoonotic campylobacteriosis, but other foods should also be considered.

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