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Epidemiology of campylobacteriosis in Denmark 2000–2015

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Summary

Campylobacter is the most frequently occurring cause of bacterial gastroenteritis in Europe. Unlike other zoonotic diseases, European-wide incidences of *Campylobacter* infections have increased during the past decade, resulting in a significant disease burden. In Denmark, campylobacteriosis is notifiable by laboratory and a unique registration system of electronic transfer and storage of notified *Campylobacter* cases linked to the national person register of age, gender and geographical location allows collection of comprehensive case data. Using national surveillance data, we describe *Campylobacter* infections in Denmark from 2000 to 2015, focusing on age-specific incidences, geography, seasonality and outbreaks. During the observed period, a total of 60,725 *Campylobacter* infections were registered with a mean annual incidence of 69.3 cases/100,000 population. From 2000 to 2014, the incidence of campylobacteriosis decreased by 20%, followed by an apparent increase of 20% from 2014 to 2015. Approximately one-third of cases were travel-related. Incidences were highest in males, young adults aged 20–29 years and children under 5 years of age. Generally, children under 10 years of age living in rural areas were at higher risk of infection. Infection patterns were seasonal with an increase from May to October, peaking in August. Outbreaks were identified each year, including four large waterborne outbreaks which all occurred following heavy rainfall events. For the most part, patterns of *Campylobacter* infection in Denmark during 2000 to 2015 remained remarkably constant and followed what is known about the disease with respect to demographic, temporal and spatial characteristics. To establish better targeted prevention and control measures, the current knowledge gaps regarding both *Campylobacter* microbiology (degree of clonal diversity and clustering) and the importance of different risk factors (food versus environment/climate) need to be filled.

KEYWORDS
Campylobacter, climate, epidemiology, outbreaks, surveillance, water-borne

1 | INTRODUCTION

Infection with *Campylobacter* spp. is one of the most widespread causes of bacterial gastroenteritis in industrialized countries, often surpassing *Salmonella* (Olson, Ethelberg, Van Pelt, & Tauxe, 2008). In 2015, the incidence of confirmed infections in Denmark was 76.6 per 100,000 inhabitants compared to 16.3 for *Salmonella* (Anonymous, 2016). This pattern is mirrored across Europe (EFSA and ECDC, 2015) as well as the USA and Australia, which also report increasing incidences during the past decade (Kaakoush, Castano-Rodriguez, Mitchell, & Man, 2015).

Most *Campylobacter* species are zoonotic with poultry, wild birds and domesticated dogs and cats as reservoirs (Mughini et al., 2014). Travelling abroad is considered one of the most important risk factors for human infection. For persons without a travel history, handling and consumption of undercooked chicken, environmental factors and contact to farm

animals are significant exposures (Domingues, Pires, Halasa, & Hald, 2012; MacDonald et al., 2015; Wingstrand et al., 2006). Water is also a vehicle for *Campylobacter* transmission, and several outbreaks have been linked to contaminated drinking water (Bartholomew, Brunton, Mitchell, Williamson, & Gilpin, 2014; Braaye, Schrijver, Wollants, van Ranst, & Verhaegen, 2015; DeFraites et al., 2014; Gubbels et al., 2012; Guzman-Herrador et al., 2015; Taylor et al., 2013; Unicomb, Fullerton, Kirk, & Stafford, 2009). Little is known about possible environmental sources, but sand, soil and water contaminated by animal faeces are all likely to play a role making outdoor activities potential risk factors – as also confirmed by *Campylobacter* outbreaks linked to hiking, mountain biking and water sports (Dale, Kirk, Sinclair, Hall, & Leder, 2010; Harder-Lauridsen, Kuhn, Erichsen, Mølbak, & Ethelberg, 2013; Stuart et al., 2010; Zeigler et al., 2014).

Campylobacteriosis is characterized by acute, self-limiting watery or bloody diarrhoea. In rare cases, infection can trigger Guillain-Barré syndrome, a paralysis-causing disorder of the peripheral nervous system. Although infection occurs in all age groups, incidences are higher in persons younger than 30 years and particularly in children (Nielsen, Ejlertsen, Engberg, & Nielsen, 2013). Acquired protective immunity likely plays an important role in *Campylobacter* epidemiology – the concept being that children are repeatedly exposed to the bacterium, through food and the immediate environment, subsequently developing partial immunity to disease, which allows them to remain asymptomatic (following most exposures) if infected as adults (Havelaar et al., 2009).

In this paper, we describe the demographic and geographical determinants of campylobacteriosis in Denmark from 2000 to 2015, summarizing the current knowledge and providing an informed basis for future infection prevention measures and disease management guidelines.

Impacts

- Incidence of campylobacteriosis is significantly higher in males, children under 5 years of age and young adults aged 20–29 years. Children living in rural areas are more likely to become infected with *Campylobacter* than children living in urban areas.
- All recent water-borne outbreaks of *Campylobacter* in Denmark occurred following unusually heavy rainfall, suggesting that predicted climate changes are an area of concern with respect to *Campylobacter* epidemiology.
- In spite of intensive control programmes targeting the broiler industry, the incidence of Danish campylobacteriosis has remained remarkably stable at high levels during the 16 years of the study, highlighting not only a potentially important role for reservoirs other than poultry but also the impact of the recent changes and improvements in the Danish *Campylobacter* notification system.

2 | MATERIALS AND METHODS

2.1 | Laboratories and registers

Diagnosis of campylobacteriosis in Denmark is undertaken at publicly financed clinical microbiology laboratories. On 1 January 2007, five geographical regions replaced the previous 16 counties in Denmark as the result of a government reform, which led to a decrease in the number of clinical microbiology laboratories nationwide, from 15 to the present 10. Most diagnostic tasks are undertaken at these laboratories from where information on notifiable diseases is sent to Statens Serum Institut (SSI, the Danish national institute for infectious disease control). The geographical uptake area for each microbiology laboratory is difficult to define; however, in our analyses we have used the borders of the surrounding municipalities. In all geographical analyses, any confounding effect of unequal activity levels between different laboratories was adjusted for.

Detection of human *Campylobacter* is mandatorily notifiable in Denmark. Since 2000, microbiology laboratories have been obliged to report a human *Campylobacter* find to SSI using paper forms. In late 2014, this reporting was supplemented by automatic data transfer from MiBa, the national database for microbiological analysis results (Voldstedlund, Haarh, & Mølbak, 2014).

Data on *Campylobacter* infections are electronically stored in the Danish Register on Enteric Pathogens (TBR) and include the unique Central Person Register (CPR) number for each patient, date of sample, name of microbiological laboratory and species determination if available. In some cases, information on travel abroad prior to disease onset (collected by the patient's GP) is also included; however, this is not mandatory and therefore generally incomplete. Further, GPs do not use a universal definition of the timing for "prior to disease onset" and thus this information refers to presumed rather than established facts. All Danish residents are listed in the Civil Registration System by their CPR number, and registrations contain information on date of birth, sex, address, municipality and region of residence for each person.

2.2 | Geographical information

At present, there are 98 municipalities in Denmark. Using a combination of socio-economic indicators, these municipalities have been classified into distant (16), rural (30), intermediate (17) and urban (35) (Sørensen, 2014). Following this classification, the residency of each patient was assigned into one of the four categories. For each municipality, an average population size during 2000–2015 was calculated using demographic information from Statistics Denmark (www.statistikbanken.dk).

2.3 | Data sources

For the general descriptive epidemiology, *Campylobacter* infections notified in Denmark from 1 January 2000 until 31 December 2015 for persons with intact CPR numbers were extracted from TBR. Danish *Campylobacter* outbreaks are detected either by an increase in case numbers in TBR or by direct notification of unusually many ill persons to local GPs or the local food authority. All food- or waterborne outbreaks are registered in a national database, FUD. Data on confirmed *Campylobacter* outbreaks between 2005 and 2015 were extracted from this database.

FIGURE 1 Mean annual incidence of campylobacteriosis in Denmark, 2000–2015

The detailed analysis of established travel-related *Campylobacter* infections included cases notified between 1 January 2008 and 31 December 2015 by Aalborg and Odense microbiology laboratories. This subset was chosen as all cases diagnosed by the two laboratories during that period were attempted to be individually interviewed about their travel activities before onset of symptoms (Ethelberg & Mølbak, 2011) rather than relying on the incomplete and biased travel information supplied by GPs along with the general case notifications.

Accumulated and age-specific population sizes per year were obtained from Statistikbanken Denmark (www.statistikbanken.dk).

Data were analysed by descriptive statistics, Poisson regression, ANOVA and two-tailed *t* tests using the STATA™ software 11.0 (Statacorp., Lakeway, TX, USA). Maps were generated using QGIS 2.14.1. (QuantumGIS, Open Source Geospatial Federation Project, www.qgis.org).

3 | RESULTS

3.1 | General demography

During 2000–2015, 60,725 notifications of *Campylobacter* infection were registered in TBR. Of these, 7,813 infections (12.9%) were registered by the patient's GP as presumably having been acquired abroad.

The overall mean annual incidence of campylobacteriosis was 69.3 cases/100,000 population ranging from 58.9 in 2006 to 84.4 in 2001 (Figure 1). From 2000 to 2014, a significant (20%) decrease in incidence was observed from 75.9 to 67.9 ($p < .001$), primarily driven by domestic cases. From 2014 to 2015, the overall reported incidence increased by 20% to 76.8/100,000 population (Figure 1). During 2004–2015, the incidence of presumed travel-related cases increased significantly ($p < .001$) from 5.9 to 12.3/100,000 population.

The overall mean incidence over time was significantly higher in males (73.2/100,000) compared to females (64.5/100,000) with an incidence rate ratio of 1.14 ($p < .01$). For domestic cases, three age groups had a significantly higher incidence compared to all other age groups ($p < .001$): 20–24 years (136/100,000), 25–29 years (114/100,000) and 0–4 years (96.8/100,000) (Figure 2). This was the same for both males and females with the exception of 0–4-year-old girls where the incidence was the same as other age groups. For presumed travel-related cases, only young adults (20–29 years) had a higher incidence compared to the rest of the population (Figure 2). Over time, domestic incidence decreased significantly for all ages between 0 and 39 years ($p < .001$) and increased for persons aged 45–54 years and over 60 years ($p < .001$). Presumed travel-related incidences increased significantly in persons aged 15–19 years and in all persons aged 35 and older ($p < .001$).

3.2 | Geography

Geographical location at municipality level was available for 94% of cases (49,737 persons) with no known travel history before disease onset. The mean annual incidence per municipality ranged from 10.4 to 86.4/100,000 population (Figure 3). Incidences of campylobacteriosis were significantly higher in intermediate and urban areas (both before and after adjustment for clinical microbiological laboratory) for the population as a whole and specifically for persons aged 50 and older while children younger than 10 years living in distant and rural areas had significantly higher incidences compared to those living in urban or intermediate areas (Table 1).

3.3 | Seasonality

A pronounced seasonality was observed for *Campylobacter* infections during 2000–2015 for both domestic and travel-related cases (Figure 4). Domestic cases increased from May to October, reaching a peak in August, while presumed travel-related cases remained higher in the beginning of the year, increasing slightly in March/April and thereafter in July, peaking in August. The seasonality of both domestic and travel-related cases was independent of year of notification as well as age, and it did not differ between rural and urban areas.

3.4 | Outbreaks

A total of 29 outbreaks involving 1823 persons were identified from 2005 to 2015 (Table 2). While a microbiologically confirmed vehicle was often not found, the most likely source was instead identified through

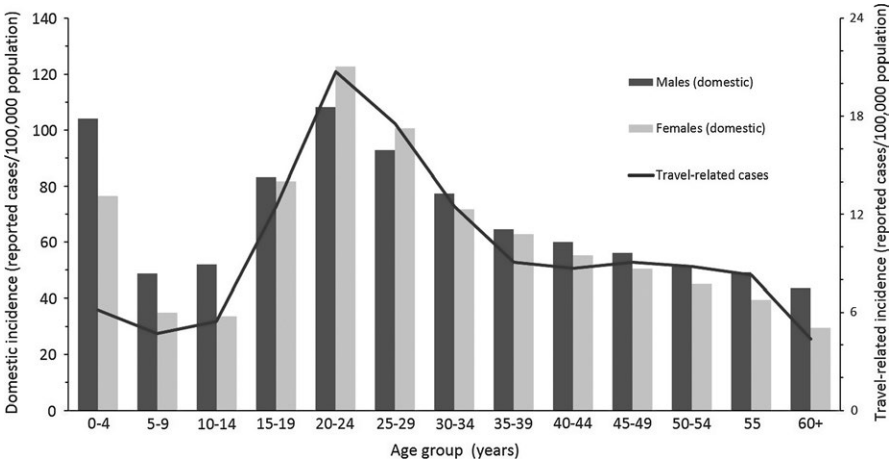

FIGURE 2 Mean age-specific incidence of domestic and travel-related campylobacteriosis in Denmark, 2000–2015

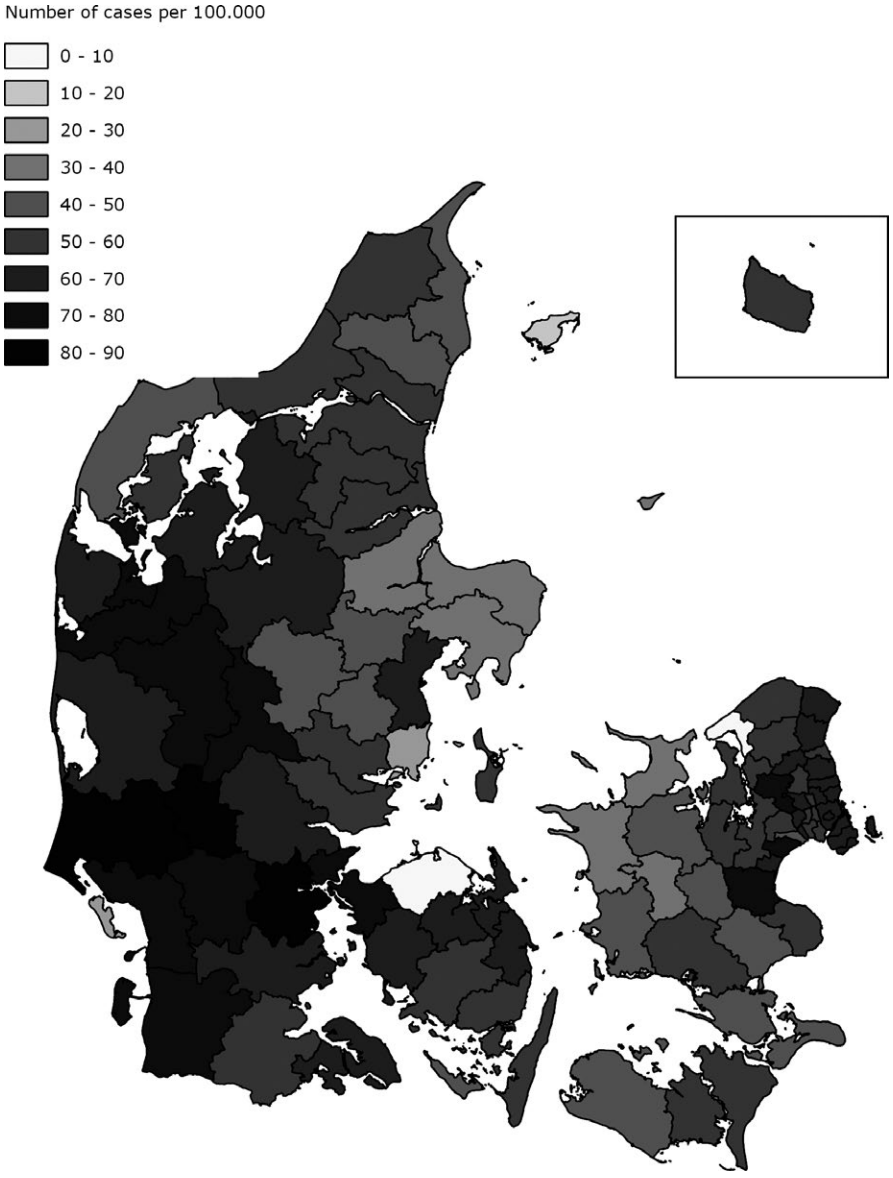

FIGURE 3 Mean incidence of *Campylobacter* at municipality level in Denmark, 2000–2015

descriptive or analytical epidemiology. Most outbreaks were of smaller scale, confined to canteens or restaurants, and generally involved chicken. During the study period, four large community-wide outbreaks occurred involving at least 1,300 persons. For three of these outbreaks, the source was confirmed to be drinking water while the fourth was linked to ingestion of sea water during a swimming competition.

3.5 | Travel

The subset of cases used for detailed analysis of travel-related infections included a total of 5,396 notifications of which 1,334 (25%) were not available for interviewing. Of the remaining 4,062 cases, 2,665 (66%) reported no foreign travel in the 14 days prior to symptom onset, while 1,393 patients (34%) had travelled abroad during this time. Patients who reported travel had visited 116 different countries. Turkey (5%), Spain (3%), Thailand (3%), India (2%) and Bulgaria (1%) were the most frequently named.

The proportion of travel-related cases ranged from 30% to 36% of all interviewed persons during 2008 to 2015 and was highest in persons aged 15–34 years (44%) and lowest in children under 10 years (5%). Generally, the risk of travel-acquired *Campylobacter* increased significantly with age ($p < .001$).

TABLE 1 *Campylobacter* in Danish municipalities

Municipality class	Incidence all ages[a]	Incidence 0–10 years[a]	Incidence 50+ years[a]
Distant	55.7	142.0*	51.6
Rural	56.5	225.6*	46.3
Intermediate	59.9	80.8	106.4*
Urban	61.2	123.9	149.5*
All municipality classes	58.3	143.1	88.5

[a]Mean annual incidence (cases/100,000 population).
*Significantly higher incidence compared to other municipality classes ($p < .001$).

4 | DISCUSSION

This paper analyses surveillance data on Danish campylobacteriosis over 16 years, from 2000 until 2015. During this time, the overall incidence was almost 70 cases/100,000 population. A significant decrease was observed in the incidence in the first 15 years, followed by a 20% increase from 2014 to 2015. This increase probably represents a notification artefact mainly caused by the increased shift towards direct electronic notification of *Campylobacter* infections (suggesting

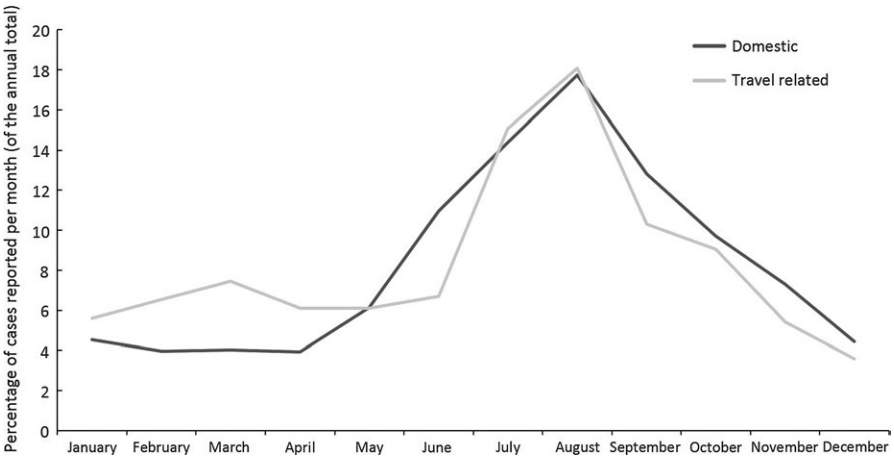

FIGURE 4 Seasonal distribution of campylobacteriosis in Denmark, 2000–2015

TABLE 2 Summary of *Campylobacter* outbreaks, Denmark 2005–2015

Year	No. of outbreaks	Total persons involved	No. persons involved in largest outbreak	Setting for largest outbreak	Vehicle for largest outbreak[a]
2005	2	68	58	Canteen	Chicken salad
2006	2	34	23	Canteen	Buffet meals
2007	7	169	140	Town	Drinking water[a]
2008	2	33	28	Tourist party	Chicken
2009	3	506	500	Town	Drinking water[a]
2010	5	846	400[b]	Town, athletic competition	Drinking water[a], Sea water[a]
2011	1	14	14	Scout camp	No suspected vehicle identified
2012	3	31	13	Canteen	Broiler meat
2013	2	7	4	Regional	No suspected vehicle identified
2014	2	5	3	Restaurant	Chicken
2015	1	110	110	Canteen (delivery to companies)	Lettuce

[a]Indicates that the vehicle was confirmed by a combination of microbiological and epidemiological evidence.
[b]Two outbreaks, each involving 400 persons.

a previous under-reporting of *Campylobacter*) as well as the recent introduction of the more sensitive diagnostic method of direct PCR on faecal matter. The decrease observed from 2000 to 2014 coincides with the implementation of the national strategy to control *Campylobacter* in broilers and retail food initiated in 2003 and including hygiene reinforcements, bonus for *Campylobacter*-negative broiler flocks, prevention of flock colonization and channelling of positive flocks to frozen production (Rosenquist et al., 2009). From 2002 to 2007, 2011–2013 and 2014–2015, the percentage of *Campylobacter*-positive broiler flocks decreased by 16%, 9.4% and 29%, respectively, and positive samples of chilled broiler meat decreased by 10%, 28% and 36%, respectively (Anonymous, 2016; Rosenquist et al., 2009). Although these reductions are significant, it is difficult to measure to which degree they directly caused a decrease in the number of human cases.

The country-to-country variations observed for campylobacteriosis across the EU is heavily influenced by differences in surveillance, diagnostic and notification practices but potentially also reflects variations in exposure to risk factors. The overall incidence of *Campylobacter* in Denmark during 2000–2015 was marginally lower than the EU average of 71 cases/100,000 population (EFSA and ECDC, 2015); however, in 6 of the 16 years, the reported incidence surpassed this average at 73–84/100,000 cases population. As for other comparatively mild gastrointestinal infections, the true incidence will be substantially higher than the number diagnosed and reported; thus in Denmark, the true incidence has been estimated at 29 times higher than that reported (Haagsma et al., 2013). This fact has also been highlighted in recent sero-epidemiological studies across Europe, which indicate infection pressures at much higher rates than reported through notification systems (Emborg et al., 2015; Teunis et al., 2013).

In our study, we found that males had overall higher infection rates than females and that incidences were higher in individuals younger than 30 years, particularly in small boys and young adults aged 20–24. The surplus of infections in males generally aligns with previous observations in other European countries (Schielke, Rosner, & Stark, 2014). Increased infection rates among children have also been demonstrated for other zoonotic bacteria (Centers for Disease Control and Prevention (CDC), 2013; Fletcher et al., 2015), which highlights both the theory of partial immunity and more frequent exposure to risk factors among children. The higher *Campylobacter* incidences in young adults could be caused by them leaving home and learning to cook for themselves, potentially slacking on kitchen hygiene ("the secondary weaning phase"). A further reason for this pattern may be the higher rates of foreign travel observed for this group in the present study and also reported previously (Simonsen, Frisch, & Ethelberg, 2008).

There was a lack of reliable travel information for a large part of the reported cases; however, active surveillance provided reliable travel information for a proportion of cases and, inferring from this, roughly a third of all Danish *Campylobacter* cases are acquired abroad which corresponds well with observations from other countries (Guzman-Herrador, Vold, & Nygård, 2012; Herbinger et al., 2016; Mughini et al., 2014; Ricotta et al., 2014). Analysis of our "travel information cohort" showed a pattern of infections acquired primarily in Asia and Southern and Eastern Europe. Interestingly, the number of infections acquired in Eastern Europe (primarily Bulgaria) increased steadily from 2008 onwards, most likely corresponding with the increased popularity and availability of tourist destinations in this region. Travel-related incidence was highest in persons aged 20–34 years, mirroring other findings (Schielke et al., 2014) and indicating a possible focus point for future information campaigns on travellers' diarrhoea.

In this study, the overall incidence of *Campylobacter* was significantly higher in urban and semi-urban areas compared to rural areas. Specifically, children living in rural areas were more frequently affected than children from urban regions and vice versa for persons aged 55 years and older. This pattern has been found previously for *Campylobacter* in Denmark (Ethelberg, Simonsen, Gerner-Smidt, Olsen, & Mølbak, 2005) as well as throughout the world (Banmali, Fritschi, & Atrie, 2006; Levesque et al., 2013; Nichols, Richardson, Sheppard, Lane, & Sarran, 2012; Schielke et al., 2014), supporting the hypothesis that children in rural areas are more frequently exposed to environmental sources of infection such as farm animals, contaminated water and wildfowl (Doorduyn et al., 2010; MacDonald et al., 2015). Unequal activity levels (i.e., not proportional to the size of the population served) of the different laboratories represented a major potential source of error in the geographical analyses. However, our analyses showed that this did not significantly impact the results.

Campylobacteriosis in Denmark from 2000 to 2015 exhibited a marked seasonal pattern with increasing incidences from May to October, peaking in August. Strong seasonal variation has often been demonstrated for *Campylobacter* infections (Jore et al., 2010; Kovats et al., 2005; Schielke et al., 2014; Strachan et al., 2013). This may be due to increased exposure because of different eating habits and changes in hygiene measures as well as more frequent contact to environmental risk factors in warmer weather. Infection rates in broiler flocks and *Campylobacter*-positive broiler meat fluctuate with the seasons, also reaching peak levels in the summer months (Anonymous, 2009; Boysen, Vigre, & Rosenquist, 2011; Meldrum, Smith, & Wilson, 2006) and creating a likely scenario for concurrent increases in human cases. Seasonal increases in presumed travel-related infections, though also apparent in the summer, were shorter in duration and corresponded with the high season for travelling in July and August. The smaller peak of presumed travel-related infections observed in March/April could reflect travel activities during the Easter holidays.

During 11 years of the study period, 29 *Campylobacter* outbreaks were confirmed. Most were linked to either direct consumption of chicken or ready-to-eat foods which, during preparation, had been contaminated by meat juice from raw chicken (Lewis, Mazick, Mølbak, Ethelberg, & Lisby, 2007; Mazick, Ethelberg, Nielsen, Mølbak, & Lisby, 2006). Three large outbreaks were caused by contamination of drinking water (Gubbels et al., 2012; Kuhn et al., 2016; Vestergaard et al., 2007); in two cases, a heavy rainfall prior to the outbreak caused a backflow of sewage water into drinking water reservoirs while the third outbreak was due to a technical error causing drinking water to become polluted by raw sewage. The latter was characterized by a mixed aetiology (*Campylobacter*, *Escherichia coli*, norovirus and several parasites), while the other two outbreaks involved only *Campylobacter*.

The fourth large outbreak was again of mixed aetiology but with confirmed *Campylobacter* stool culture from several cases. It was linked to an unusually heavy rainfall over Copenhagen, which, one day prior to a triathlete competition, resulted in sewer overflow and a severe sewage pollution of the sea water in which the athletes swam (Harder-Lauridsen et al., 2013). Studies of the geographical distribution of *Campylobacter* cases in Denmark have suggested that contaminated drinking water could also play a role for the sporadic cases (Ethelberg et al., 2005; Jepsen, Simonsen, & Ethelberg, 2009).

For more than a decade, campylobacteriosis has been the most prevalent bacterial gastrointestinal illness in Europe. Here, we demonstrate that the disease in Denmark from 2000 to 2015 mostly followed known patterns with respect to demography, seasonality and geography. We confirm that young adults, particularly boys, and children living in rural areas are at significantly higher risk of infection. Interestingly, all waterborne *Campylobacter* outbreaks occurred immediately following a heavy rainfall, highlighting a potential area of future concern as heavy precipitation events are predicted to become more frequent (Kovats et al., 2014). The number of cases overall was surprisingly stable at high levels over the study period and intervention programmes, specifically targeting the broiler production industry, have at the most been only partly effective. This can be due to a multitude of factors such as the intervention programmes alone being insufficient, that other transmission routes play important roles and not least that changes and improvements to the surveillance system have allowed a continuously high degree of case detection. Generally, it remains a fact that there are still a number of unknowns concerning the exact risk factors underlying campylobacteriosis transmission patterns. The road towards a better understanding hereof may lie in new tools available to us: next-generation sequencing which enables high resolution and very accurate analysis of bacterial strains, identifying the extent of single-strain human clustering (mini outbreaks) and evaluating the diversity of veterinary and human clonal populations. The combination of these detailed typing methods and the increased availability of electronically stored data could pave the way for a future with improved knowledge about *Campylobacter* with respect to identification of temporal and geographical shifts, sources of transmission and infection control.

CONFLICTS OF INTEREST

None to declare.

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