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Original article

A foodborne outbreak of campylobacteriosis at a wedding – Melbourne, Australia, 2022

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Abstract

Campylobacter is the most common bacterial cause of foodborne gastroenteritis in Australia; however, outbreaks caused by the pathogen are relatively uncommon. In March 2022, the Victorian Department of Health was notified of a gastrointestinal illness in 20 guests following attendance at a wedding reception. Two of these individuals were notified with laboratory-confirmed campylobacteriosis, and an investigation was undertaken to identify the source of the infection and implement strategies to prevent further illness.

A case-control study was conducted to determine the likely source of infection. Cases were defined as attendees of the wedding reception, with onset of diarrhoea and/or abdominal cramping 1–10 days after attending the function. Controls were randomly selected from the remaining list of non-ill guests. Cases and controls were interviewed using a standardised, menu-based questionnaire. Food preparation processes were documented, and food samples collected.

A total of 29 wedding guests met the case definition. Cases reported onset of illness 2–5 days following the wedding and major symptoms included abdominal cramping (100%), diarrhoea (90%), headache (79%), and fever (62%). Two cases were hospitalised, one with ongoing secondary neurological sequelae. Illness was significantly associated with consumption of a duck breast brioche canapé containing duck liver parfait (odds ratio = 2.85; 95% confidence interval: 1.03–7.86). No leftover food samples were available for testing.

The investigation found that the duck canapé was the likely vehicle of infection. Consistent with the literature on *Campylobacter* transmission, it is likely that inadequate cooking of the duck liver for the parfait was the contributing factor that led to illness. This highlights the risks posed by undercooked poultry dishes, and shows that education of food handlers remains a priority.

Keywords: *Campylobacter jejuni*; campylobacteriosis; foodborne illness; gastroenteritis; duck liver; outbreak investigation

Introduction

Campylobacter spp. are the cause of the most common notifiable bacterial infection causing gastroenteritis in Australia, and a leading cause of gastroenteritis worldwide. In 2019, there were 35,869 cases reported in Australia, with a national notification rate of 143.5 per 100,000;[1] however, this likely only represents the tip of the iceberg of the true number of cases that occur annually.[2] Despite the high incidence of the disease, most infections are sporadic, and outbreaks of campylobacteriosis remain relatively rare. Between 2001 and 2016, 84 outbreaks were reported in Australia, with 61% (n = 51) attributable to foodborne transmission. Of those outbreaks with an identified food vehicle, poultry meat or offal was implicated in the majority (n = 28; 85%), with liver dishes such as paté contributing to a significant proportion (n = 11; 39%).[3]

The incubation period for *Campylobacter* is generally two to five days (range 1–10 days).[4] Illness is often characterised by diarrhoea that can be bloody; fever; and abdominal cramping that can last for one to two weeks.[5] The disease can also result in secondary postinfectious complications, including reactive arthritis (2–5%); irritable bowel syndrome (9–13%); and Guillain-Barré syndrome (0.1%), a secondary autoimmune complication that can result in neurological symptoms such as paralysis.[6]

On 4 March 2022, the Victorian Department of Health received a notification from council that 20 out of 212 guests had experienced symptoms of gastrointestinal illness following attendance at a wedding function in mid-February. Subsequent to the initial complaint, two laboratory-confirmed campylobacteriosis cases were identified to have attended the wedding, and an investigation was commenced. This report details the investigation undertaken to determine the potential source of the illness and describes the public health actions taken to prevent further illness.

Methods

Epidemiological investigation

A retrospective case control study was performed. The study was conducted as an outbreak investigation under the Victorian *Public Health and Wellbeing Act 2008*, so ethics approval was not required.

A confirmed case was defined as an individual who had attended the wedding function, had an onset of diarrhoea and/or abdominal cramping within the following ten days, and had a laboratory-confirmed faecal sample for *Campylobacter* spp. A probable case was defined as an individual who had attended the wedding function and had an onset of diarrhoea and/or abdominal cramping within the following 10 days.

Unmatched controls were selected by randomising the list of non-ill guests provided by the wedding organisers. An attempt was made to interview two controls for every case. Controls were reclassified as cases if they reported symptoms fitting the case definition. Individuals were excluded from being controls if they reported symptoms of gastrointestinal illness in the ten days following attendance at the wedding, if they did not wish to provide contact details to the Department, or if they were lost to follow-up.

Cases and controls were interviewed over the phone using a structured menu-based questionnaire. It collected demographic and clinical information, toilet use, and food and beverage consumption. Participants were contacted a maximum of three times, after which they were considered lost to follow-up. Any participants who were contacted but declined to be interviewed, or who had a disconnected phone number, were also considered lost to follow-up.

Statistical analysis

Data were analysed using Stata v15.4. Descriptive analysis was performed on the demographic and clinical variables. Probability of difference in sex and age between cases and controls was determined using Chi square test and Wilcoxon rank sum test respectively.

Univariate analysis was conducted to determine crude odds ratios (OR) and 95% confidence intervals (CI) for association between food or environmental exposures and illness, and the Chi square test or Fishers exact test (for a cell count < 5) was used to determine statistical significance ($p < 0.05$). Where a food exposure contained a cell count of zero, exact logistic regression was used to calculate adjusted odds ratios (aOR). Food exposures significantly associated with illness following the univariate analysis were stratified and/or put into a multivariate logistic regression model to account for the effects of confounding and an adjusted odds ratio determined with 95% CI and statistical significance ($p < 0.05$).

Environmental & microbiological investigation

Local government environmental health officers (EHOs) inspected three premises managed by the catering group that provided food for the wedding. These comprised the outbreak venue; the central production kitchen (where most food had been prepared); and the premises where the duck liver paté was manufactured and supplied to use for the parfait in the duck canapé. Food storage and preparation areas were examined, and information was collected on food preparation processes and staff illness.

Human faecal specimens and food samples were collected and forwarded to private pathology companies or to the Microbiological Diagnostic Unit Public Health Laboratory (MDU PHL) for analysis.

A sample of a different batch of duck and cherry paté log used in the duck canapé was collected from the supplier of the duck paté and sent for analysis, as were samples of four other high-risk foods manufactured on site.

Results

Epidemiological investigation

Descriptive epidemiology

Eighty-five guests were interviewed. Of these, 29 individuals met the case definition (4 confirmed, 25 probable); 54 were included as controls. Two interviewed guests reported nausea but did not fit the case definition and were excluded from the study. An additional 43 individuals declined to provide contact details, and 15 were lost to follow-up.

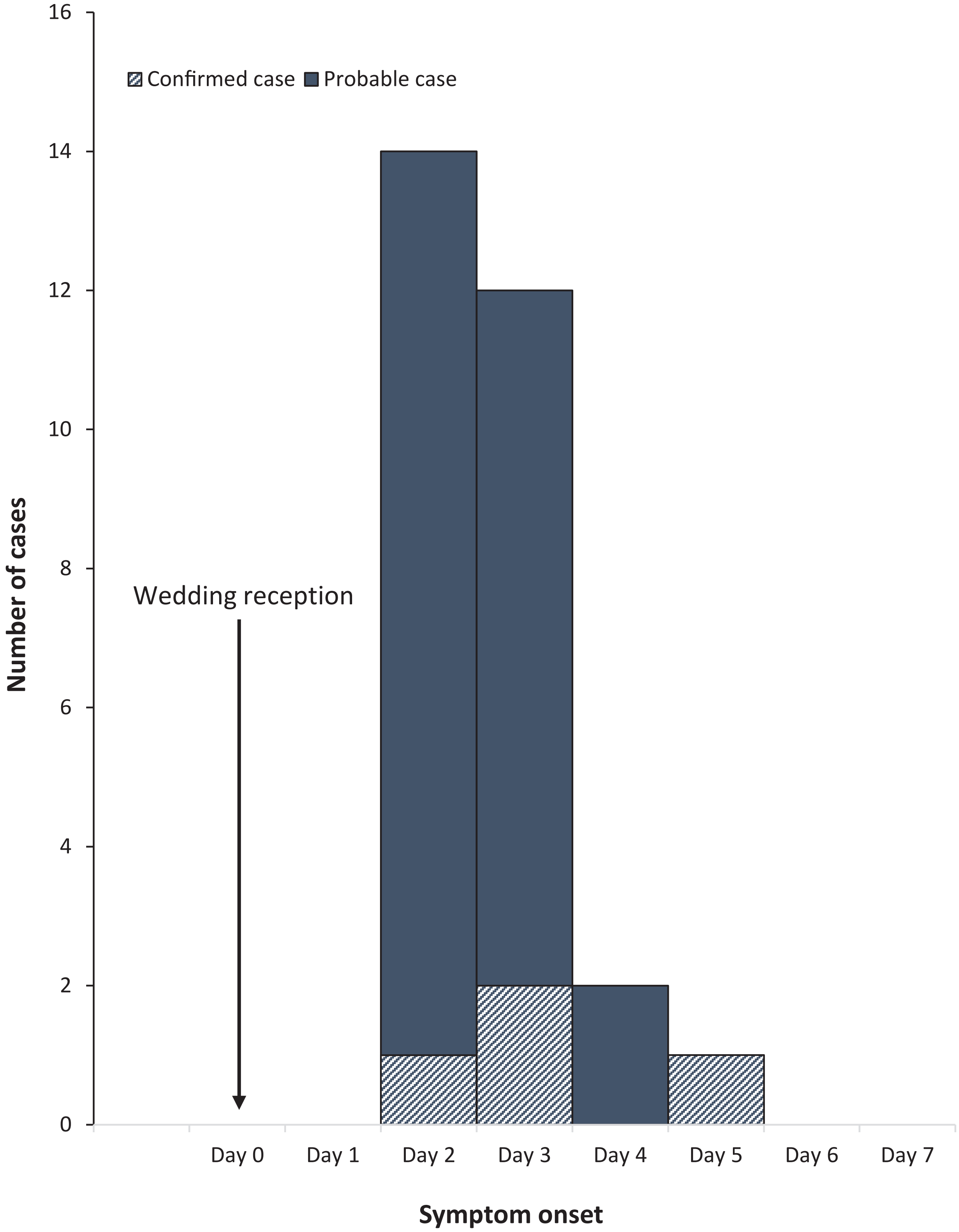
All cases reported onset of symptoms 2–5 days (median three days) after the reception (Figure 1). Demographic and symptom details for cases and controls are shown in Table 1. There was a significant difference in both sex and age distribution between the two groups, with cases more likely to be male (p = 0.001) and younger (p = 0.023) (median 29 years; range 18–69) than controls (median 45 years; range 31–71). Diarrhoea, abdominal cramping and nausea were present in nearly all cases (Table 1). Seventeen cases (59%) presented to a medical practitioner and two cases (7%) were hospitalised. While the median duration of gastrointestinal illness was six days (range 1–12 days), at time of interview two cases were experiencing ongoing secondary complications, including a probable case with irritable bowel syndrome and a probable case with neurological sequelae suspected to be Guillain-Barré syndrome.

Analytical epidemiology

Following univariate analysis, canapés were significantly associated with illness (p = 0.002), with 24 (83%) of those who experienced gastrointestinal illness consuming some type of canapé, and the odds of cases eating canapés 5.17 times more likely than controls (Table 2). Of the canapés, 14 cases (50%) ate the duck canapé (duck breast with apple relish brioche with duck liver parfait) and 15 cases (54%) ate the lamb and fetta bastilla. Both foods had elevated univariate OR (3.33 and 2.38 respectively); however, only the duck canapé was statistically significant with a 95% CI 1.11–9.97. The wagyu beef main also had an elevated OR of 2.71; 91% CI: 0.91–8.72; and p = 0.048 and was borderline significant. Several other food items had elevated OR greater than one; however, there were no statistically significant associations with consumption of these other food items and illness (data not shown).

To adjust for potential confounding of cases consuming both duck canapé and wagyu beef, stratification was performed (Table 3); the aOR for beef remained elevated, but was no longer significantly associated with illness when adjusted for eating duck (aOR: 2.25; p = 0.129). The duck canapé remained significantly associated with illness independent of the beef (aOR: 2.85; p = 0.043). When stratification was performed to control for age (Table 3), there was significant evidence of effect modification (p = 0.029), with cases aged 10–29 years 13.2 times

Figure 1: Number of confirmed and probable cases by day of symptom onset, Victoria, February 2022 (n = 29)



more likely to have eaten duck canapé than controls in the same age group, while there was no association seen in cases aged ≥ 30 years.

There was, however, some loss in the strength of association between consumption of the duck canapé and illness when adjusted for sex (Table 4). Despite the loss in significance, duck canapé still had a stronger association with illness (aOR: 2.62; $p = 0.069$) than with sex (aOR: 2.13; $p = 0.146$).

Table 1: Demographic and clinical characteristics of cases and controls, Victoria, February 2022 (n = 83)

Characteristic	Cases (N = 29)		Controls (N = 54)		*p* value
	n	%	n	%	
Age					
Median (range), years	29 (18–69)		45 (31–71)		0.023[a]
10–29	17	(59%)	13	(25%)	
30+	12	(41%)	39	(75%)	
Total	**29**		**52**[b]		
Sex					
Male	16	(55%)	16	(30%)	0.001[c]
Female	13	(45%)	38	(70%)	
Symptom					
Abdominal cramping	29	(100%)			
Diarrhoea	26	(90%)			
Nausea	25	(86%)			
Headache	23	(79%)			
Fever	18	(62%)			
Vomiting	13	(45%)			
Bloody diarrhoea	6	(23%)			
Secondary complications	2	(7%)			

a Two-sample Wilcoxon rank-sum (Mann-Whitney) test.

b Two controls had missing data.

c χ^2 test.

Environmental investigation

The paté used in the preparation of the parfait for the duck canapé was a duck and cherry paté log containing both duck (24%) and chicken livers. The poultry livers used to make the paté were obtained frozen and thawed in the fridge. The paté supplied to the wedding had been prepared more than two months prior, and no additional livers from the same batch used to make the paté were available for testing.

Livers were reportedly cooked at 120 °C until an internal temperature of more than 65 °C was reached for more than 10 minutes, blended with duck fat and butter, and rested for 20 minutes, which reportedly resulted in an internal temperature rise to 68–71 °C due to residual heat. However, no temperature records were kept or were available for the batch supplied to the wedding to confirm this process. Environmental swabs of blending and sieving equipment on site at the manufacturer's premises were also requested but not obtained.

The duck liver paté was whipped into a parfait and placed into piping bags at the central production kitchen. The premises also prepared and cooked the duck breast. Raw duck breast was marinated and refrigerated overnight at a recorded temperature of 5 °C. The next day it was washed and cold smoked for one hour, before being pan-fried to golden and roasted at 160 °C for 6–8 minutes. The finishing temperature was

Table 2: Univariate analysis of food exposures,[a] Victoria, February 2022 (n = 83)

	Cases			Controls					
Exposure	**Total**	**Exposed**	**%**	**Total**	**Exposed**	**%**	**OR[b]**	**95% CI[c]**	***p* value[d]**
Canapé – any	**29**	**24**	**82.8**	**54**	**26**	**48.2**	**5.17**	**1.58-19.57**	**0.002**
Eggplant miso bun	29	10	34.5	52	11	21.1	1.96	0.62-6.08	0.189
Duck breast canapé	**28**	**14**	**50.0**	**52**	**12**	**23.1**	**3.33**	**1.11-9.97**	**0.014**
Lamb and fetta bastilla	28	15	53.6	52	17	32.7	2.38	0.84-6.78	0.069
Mains – any[e]	29	29	100.0	54	51	94.4	2.12	0.22–∞	0.540
Wagyu beef	**29**	**22**	**75.9**	**54**	**29**	**53.7**	**2.71**	**0.91-8.72**	**0.048**

a Note: not all exposures are shown.

b OR: odds ratio.

c CI: confidence interval.

d χ^2 to determine statistical significance unless otherwise indicated ($p < 0.05$ indicates statistical significance).

e Odds ratio and confidence intervals calculated using exact logistic regression.

Table 3: Stratified analysis of food exposures and age, Victoria, February 2022 (n = 80)

Characteristic	aOR[a]	95% CI[b]	*p* value
Wagyu beef adjusted for:			
Food items			
Duck canapé	2.25	0.79–6.43	0.129[c]
Duck canapé adjusted for:			
Food items			
Wagyu beef	2.85	1.03–7.86	0.043[c]
Age group[d]			
10–29 years	13.20	1.71–149.28	
30+ years	0.81	0.07–5.22	0.029[e]

a aOR: adjusted odds ratio.

b CI: confidence interval.

c Mantel-Haenzel χ^2 test ($p < 0.05$ indicates statistical significance).

d Some data missing – analysis performed using N = 52 controls.

e Mantel-Haenzel test of homogeneity ($p < 0.05$ indicates statistical significance).

verbally reported to have been 74 °C. Following cooking, it was then blast chilled and sliced. The whipped parfait and sliced duck were then sent to the reception venue for assembly.

Several processes at the central production kitchen were ambiguous, including how the duck breast was cold smoked; whether kitchen equipment used to whip the paté was used to prepare other food items; and the details of cleaning and sanitisation protocols for the

Table 4: Association of consumption of duck canapé with illness when adjusted for confounding by sex, Victoria, February 2022 (n=80)

Characteristic	aOR[a]	95% CI[b]	*p* value[c]
Duck canapé	2.62	0.93–7.40	0.069
Male sex	2.13	0.77–5.90	0.146

a aOR: adjusted odds ratio.

b CI: confidence interval.

c Multivariate logistic regression ($p < 0.05$ indicates statistical significance).

whipping/blending equipment. Council additionally reported that all food preparation was performed on a large bench simultaneously and knives used were washed in the sink and wiped between usage, but not sanitised.

Following inspection of the reception venue, no significant issues with hygiene and food storage were identified. The venue did not cook any of the ingredients, rather only reheated and assembled pre-cooked, pre-washed and pre-prepared ingredients from the central production kitchen. It was noted, however, that staff did not use gloves when slicing and mixing ingredients, and that salads and sides were mixed by hand in stainless steel mixing bowls.

The EHOs were unable to determine whether canapé ingredients were prepared in a single batch specifically for the outbreak function, or if they were prepared and supplied to multiple functions/venues simultaneously. No complaints of illness were reported from three additional functions that had occurred on either side of the wedding reception; however, none of these events had been served duck canapé.

Laboratory investigation

Of 11 faecal samples obtained, four had *Campylobacter* isolated by bacterial culture and/or detected by polymerase chain reaction. Of these, two were further speciated and identified as *Campylobacter jejuni*. No other bacterial or viral pathogens were detected.

No leftover food samples from the reception venue were available to undergo microbiological testing. No *Campylobacter* or other bacterial pathogens were isolated from the additional food samples collected from the paté manufacturer.

Discussion

The results from the epidemiological investigation support the hypothesis that the duck breast with apple relish brioche canapé was the most likely vehicle of infection in this outbreak.

The duck canapé remained the only food with elevated OR that was significantly associated with illness following univariate analysis and stratification, with cases 2.85 times more likely to have eaten duck canapé than controls. There was strong evidence of an age-dependent effect, with 10–29-year-old cases 13.2 times more likely to have eaten the duck canapé. It's important to note, however, that when adjusted OR were calculated to control for confounding by sex, there was a loss in strength of association of the duck canapé with illness, likely a result of the small sample size.

The canapé contained both duck breast meat and duck liver parfait, both of which fit the biological plausibility of the source of transmission. *Campylobacter* is a commensal organism in the gastrointestinal tract of poultry and is also a frequent internal and external contaminant of poultry liver.[7] The ability of the liver to concentrate the pathogen and provide optimal growth conditions, with a neutral pH and high water activity, supports the rationale that the liver parfait in this outbreak could be considered the higher risk food component in the canapé, rather than the duck breast.

Additionally, *Campylobacter* contamination of chicken meat has been shown to be more commonly on the skin of the bird than permeating into the muscle.[8] If a similar circumstance holds for duck meat, pan searing the duck breast followed by roasting to an internal temperature of 74 °C would have been sufficient to destroy any bacteria contaminating the meat.

Prevalence of *Campylobacter* in poultry liver has been reported to be as high as 90–96% in retailed livers from Australia and New Zealand.[9,10] In addition, paté and parfait have been linked to a number of outbreaks of campylobacteriosis both internationally and within Australia.[3,11–16] The majority occurred in restaurant or commercial catering settings.[3]

In the majority of paté- and liver-associated outbreaks, inadequate cooking or undercooking of the liver is thought to be the significant contributing factor. This has included shallow frying or lightly cooking to retain the pink colour;[11,13] cooking only to a core temperature of 60 °C;[17] or not holding at adequate temperature for long enough.[18] Inactivation of *Campylobacter* has been shown to be proportional to cooking time.[4] It has also been shown that pan-frying does not always uniformly heat all livers to the desired core temperature.[19]

Following a review of outbreaks linked to poultry liver,[15] guidelines have been developed by Food Standards Australia (FSA) recommending that whole livers are cooked to an internal temperature of 70 °C for at least two minutes, measured using a digital probe.[20] The guidelines also suggest that the safest way to prepare paté is to bake the whole dish in a waterbath at temperatures greater than 150 °C for two hours. These methods ensure the final paté product reaches an internal temperature sufficient to kill pathogenic bacteria.

It is unclear if the livers were cooked in a waterbath or pan fried, and whether internal temperatures reported were reached, as no temperature records were kept. If prepared in a waterbath, temperatures and cooking times were likely insufficient, based on the FSA guidelines; and if pan-fried, uneven cooking may have resulted in some livers failing to reach an appropriate internal temperature.

Neither the duck breast nor the liver parfait was a component of any other dish; however, 14 cases (50%) reported not eating duck canapé and still became unwell. While this may be attributed to misclassification of food exposures, it may also be the result of cross-contamination during food preparation. While there were reports of staff not wearing gloves and mixing dishes by hand at the wedding venue, the risk of cross-contamination was likely higher at the central production kitchen where equipment used to whip the paté into parfait may have been used on other ready-to-eat foods. Raw duck breast was also washed prior to cooking, which may have contaminated the sink and other surfaces. In addition, all food products were reportedly prepared together on a single table, and knives were not properly sanitised between usage. While biological plausibility and the univariate analysis implicate the duck canapé as the likely source of illness, the small study sample size and lack of microbiological evidence mean cross-contamination of other foods cannot be ruled out.

Conversely, 12 controls (22%) reported eating the duck and didn't become unwell. This may be attributable to multiple batches of canapés being prepared and served, some which were not contaminated or which may have had uneven distribution of *Campylobacter* within the batch. Additionally, it could be related to dose response or prior immunity. While the infectious dose of *Campylobacter* is low—less than 500 organisms can cause infection[6]—the dose required to produce symptomatic illness may be higher, depending on *Campylobacter* strain virulence and immune status of the host.[21] There is evidence to suggest that prior infection with *Campylobacter* provides acquired immunity which confers protection against symptomatic infection, and that seroprevalence to *Campylobacter* increases with age, leading to a reduction in symptomatic infection despite repeat exposures.[22]

Limitations

A major limitation of this investigation was the inability to obtain microbiological evidence of *Campylobacter* in any food samples. Instead, evidence obtained from the environmental investigation, as well as the large body of evidence in the literature reporting an association with *Campylobacter* infection in humans and consumption of poultry meat and poultry liver, support the results obtained from the epidemiological investigation.

The small sample size resulted in a lack of statistical power, making it difficult to obtain a discernibly high OR for any single food exposure. Analytical studies in outbreak investigations are almost always limited by statistical power, as there are constraints on the number of cases able to be included.

While use of unmatched controls led to significant differences in age and sex between cases and controls, we attempted to control for confounding by these variables during analysis using stratification and multivariate logistic regression. Selection bias was curtailed as much as possible by randomising controls from the same population that generated the cases. To limit recall bias, a standardised questionnaire was used, listing all foods on the menu with no open-ended questions; however, the delay between the event and notification of gastrointestinal illness may have had an effect on recall of foods eaten.

Conclusion

The evidence suggests this was a foodborne outbreak caused by *Campylobacter jejuni*, with the duck and apple relish brioche canapé the most likely food vehicle. Consistent with the literature on *Campylobacter* transmission, it is likely that inadequate cooking of duck liver for the parfait was the contributing factor that led to illness. This emphasises risks posed by poultry dishes if undercooked, and shows that education of food handlers in preparing poultry liver remains a priority. It also highlights the need to ensure food safety procedures are properly monitored and reviewed to ensure adherence.

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