



A large scale waterborne *Campylobacteriosis* outbreak, Havelock North, New Zealand

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SUMMARY

Background: We describe the investigation of a *Campylobacter* outbreak linked to contamination of an untreated, groundwater derived drinking water supply.

Methods: We analysed epidemiological data collected from clinician-confirmed diarrheal cases and estimated the total burden of Havelock North cases using an age-adjusted cross-sectional telephone survey. *Campylobacter* isolates from case fecal specimens, groundwater samples, and sheep fecal specimens from paddocks adjacent to the drinking water source were whole genome sequenced.

Findings: We estimate between 6260 and 8320 cases of illness including up to 2230 who lived outside the reticulation area, were linked to the contaminated water supply. Of these, 953 cases were physician reported, 42 were hospitalized, three developed Guillain-Barré syndrome, and *Campylobacter* infection contributed to at least four deaths. Of the 12 genotypes observed in cases, four were also observed in water, three were also observed in sheep and one was also observed in both water and sheep.

Interpretation: The contamination of the untreated reticulated water supply occurred following a very heavy rainfall event which caused drainage of sheep feces into a shallow aquifer. The existence of a routine clinical surveillance system for campylobacteriosis facilitated identification of the outbreak, recovery of clinical isolates, and early testing of the water for pathogens. Genotyping of the *Campylobacter jejuni* helped define the source of the outbreak and confirm outbreak periods and cases. Expected increases in heavy rainfall events and intensification of agriculture mean that additional safeguards are needed to protect populations from such drinking water outbreaks.

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Research in context

Evidence before this study

We searched PubMed and Web of Science with the keywords "Water" and "Outbreak" for papers published in any language up to November 30, 2019. We performed a backwards search of the reference lists of key papers for further

relevant studies, as well as searching for papers citing those papers. Comprehensive reviews were published in 2015, 2017 and 2019 with over 300 drinking water outbreaks in the industrialized world reported since 2000. Despite estimates of >100,000 people becoming ill, in many outbreaks a causative agent is not identified among clinical cases, and isolation of pathogens from contaminated water is very rarely achieved. While contamination of groundwater and surface water following heavy rainfall has been documented, the evidence to

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date has been insufficient to stimulate the necessary preventative measures.

Added value of this study

This is the largest ever recorded campylobacteriosis outbreak, emphasizing the significant disease burden that waterborne outbreaks can cause. This study provides arguably the most comprehensive characterization of a waterborne outbreak, with conclusive genetic evidence linking sources of infection (sheep on paddocks adjacent to the drinking water source) with drinking water and ultimately the clinical cases. Whole genome sequencing also augmented the epidemiologic investigation by discriminating between outbreak-linked and unlinked cases, confirming the suspected exposure period, and defining the geographical scope of the outbreak. This investigation also showed the value of cross-sectional surveys of the exposed population during the outbreak to rapidly assess the outbreak magnitude, its population impact and to support an early response.

Implications of all the available evidence

This and previous studies highlight the vulnerability of groundwater systems to contamination by surface water, and the need to consider the risk from extreme weather events which are likely to become more frequent. Source protection, suitable water treatment, regular monitoring of water, and epidemiological surveillance of disease incidence are key requirements for provision of a safe water supply. Enhancements to these activities are required following heavy rainfall or other change events, to minimize the impacts of any contamination on the water supply. Culture-independent diagnostic approaches threaten future molecular epidemiology investigations if steps are not taken to isolate causative organisms.

Introduction

Waterborne illness continues to impose a significant health burden, with documented waterborne outbreaks regularly reported in Europe, North America and elsewhere.^{1–4} *Campylobacter* has been identified as a contributing pathogen in many of these outbreaks and poses a significant risk for unchlorinated groundwater systems, due to its ability to survive several weeks in cool, wet environments and infect at low doses.^{5–7} Campylobacteriosis is the most frequently notified disease in New Zealand, with 6000–7000 cases notified each year.⁸

In August 2016 the town of Havelock North, in New Zealand, had an unchlorinated reticulated water system supplying 14,118 residents. Within 30 miles (50 km) are the neighbouring cities of Hastings (population ~64,000) and Napier (population ~61,100), which together with other small rural communities (totalling another ~15,000) comprise the Hawke's Bay District Health Board catchment area. On 12 August 2016 the Health Board was alerted to an increase in people with diarrheal illness presenting to the Hawke's Bay Hospital, elevated campylobacteriosis notifications from the region and an increase in school absenteeism. Routine microbiological testing from the Havelock North reticulated water supply taken on 11 August was positive for *E. coli*. Heavy rainfall of 6.5 inches (165 mm) during 5–6 August was suspected to have contributed to the contamination of the water supply. The water supplier (Hastings District Council) responded the same day by issuing a 'boil water notice' and initiating chlorination of the Havelock North reticulated water supply.

We describe the epidemiologic and molecular investigation into the source and magnitude of the outbreak and discuss the public health response initiated to prevent similar events from occurring in the future.

Methods

Epidemiologic case investigation

Data were collected and entered into the national web-based surveillance system for notifiable diseases, including positive laboratory results, demographics and risk factors (reticulated water consumption, occupation, recent overseas travel). For enteric diseases, including campylobacteriosis, cases are legally required to be reported to the local Medical Officer of Health by laboratories and health practitioners. Due to overwhelming demand on primary care services during the outbreak, physician reporting was facilitated through the provision of telephone consultations and active case finding among high-risk populations.

A probable campylobacteriosis case was defined as an individual who consumed Havelock North reticulated water between 5 and 12 August 2016 with clinician-confirmed diarrhea between 7 and 24 August 2016. A confirmed campylobacteriosis case was the same as a probable campylobacteriosis case but with a fecal specimen positive for *Campylobacter* spp. Together these probable and confirmed cases are, in this paper, referred to as outbreak cases. Unlinked cases in this paper resided in the Hawke's Bay, and meet requirements to be a confirmed or probable case except that they did not report the consumption of Havelock North reticulated water.

The duration of the outbreak was defined as the period when the daily count rate of notified *Campylobacter* infections in Hawke's Bay was greater than a threshold value calculated as the mean plus two standard deviations of three years of daily count data. The three years' data closely fitted a Poisson distribution from which the mean was calculated.

During the initial phase of the outbreak (7–16 August) clinicians were encouraged to collect fecal specimens for probable cases to identify pathogens contributing to the outbreak. On confirmation of *Campylobacter* spp. as the likely etiologic pathogen, routine surveillance was modified. Sentinel surveillance in two primary care practices serving Havelock North and adjacent areas replaced the usual testing practices for gastroenteritis cases. Fecal specimens were collected from the first seven patients meeting the case definition twice weekly during 17 August to 9 September 2016.

Confidence intervals (95% CI) for rates were calculated using binomial distribution for proportions. P-value for demographic group was calculated using Pearson's chi-squared test. Stata 15 was used for statistical analysis.

To estimate the proportion of households and residents affected by the outbreak, a cross-sectional study of Havelock North residents was also conducted by randomly surveying 660 households supplied by the Havelock North reticulated water system (conducted as three rounds of telephone surveys using the same panel of households, with the last survey on the 22 August). A case was defined as a resident of Havelock North with exposure to the Havelock North reticulated water supply during 5–12 August who developed diarrheal disease during 7–22 August. Results were age-adjusted to the population of Havelock North.

Microbiological investigation

Fecal samples were screened for a range of bacterial, viral and protozoan enteric pathogens (details in Supplementary Appendix). *Campylobacter* spp. were isolated from clinical specimens by streaking feces on mCCDA agar plates incubated microaerobically at 42 °C for 48 h, with putative *Campylobacter* isolates confirmed biochemically. Reticulated and bore water specimens were collected on 12 August, prior to chlorination, from four points within the distribution network and from sample taps at each of

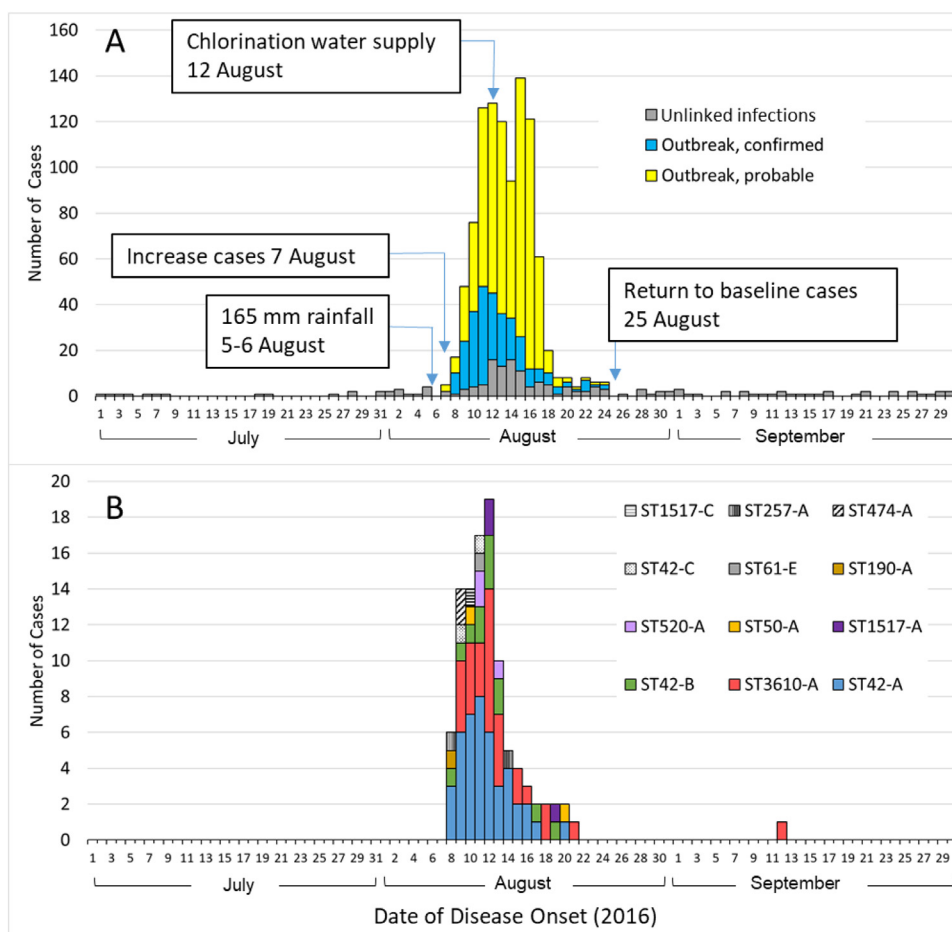


Fig. 1. Reported campylobacteriosis in the Hawke's Bay from July to September 2016 graphed according to onset of symptoms. Panel A. Confirmed, probable and unlinked reported campylobacteriosis cases. Panel B. Genotypes of outbreak related cases.

the two bores (wells) supplying the network. Additional samples from bores were collected on 19 and 24 August. Colilert (IDEXX, Westbrook, Maine, USA) assays were used to enumerate total coliforms and *E. coli*. *Campylobacter* spp. were isolated from water samples following membrane filtration of up to 1 L of water, with filters enriched in Exeter or Bolton broth and then plated onto agar plates (Supplementary Appendix).⁷ Sixteen sheep fecal samples were collected on 23 August from paddocks close to the bores, with *Campylobacter* spp. isolated from 0.1 g of sheep feces using the same enrichment procedure as for the water samples.

Isolates were whole genome sequenced using an Illumina MiSeq. Genomes were assembled, seven gene multilocus sequence type (MLST), and 1343 core genome MLST⁹ (cgMLST) allele assignments undertaken using BioNumerics 7.6 (Applied Maths, Belgium). Isolates were compared using minimum spanning trees with genomic clusters designated by the conventional seven gene MLST name, followed by a letter for isolates with fewer than three allelic differences.

Results

Epidemiologic case investigation

Post outbreak analysis of notified cases in Hawke's Bay showed a significant increase above baseline rates for cases with onset dates starting on 7 August 2016. The higher rates continued until 24 August 2016. Between these dates, 225 confirmed and 728 probable campylobacteriosis cases were notified, with the epi-

demio peak occurring during 11–16 August (Fig. 1). The highest rates were observed among those aged over 60 years (Table 1). Four deaths occurred in confirmed cases aged ≥ 70 years living in aged residential care. Three Guillain-Barré syndrome cases were identified; two occurred in Hawke's Bay residents, and one in a visitor. Other enteropathogens did not occur at rates higher than baseline levels.

Most cases ($n = 763$) lived within the Havelock North reticulated water zone, with another 96 living in Hastings, 21 in Napier, 50 in other areas of the Hawke's Bay, and 23 outside of the region (Fig. 2). Assuming an outbreak period of 7–24 August 2016, the risk of a Havelock North resident becoming a case was 29.7 (95% CI: 21.8–41.5) times greater than for residents of other Hawke's Bay towns.

All 23 cases from outside the Hawke's Bay that were included visited Havelock North during the period when the water is likely to have been contaminated, and reported drinking Havelock North water. Four of the cases residing outside Hawke's Bay had single day exposures to Havelock North water (7, 8, 10 and 12 August), and incubation periods before onset of symptoms of between 1 and 5 days. The remaining 19 cases had a possible range of 1–10 days between exposure and onset, with a median of three days.

The household cross-sectional study estimated that over one-third (39%) of residents and over half (56%) of households developed symptoms of gastroenteritis, with an age-adjusted estimate of 5540 gastroenteritis infections in approximately 14,118 residents supplied by Havelock North reticulated water (95% CI 5010–6080 infections). This corresponds to a case-ascertainment ratio of 7.3

Table 1
Demographic characteristics and clinical outcomes of confirmed and probable campylobacteriosis cases.

Demographic	Cases	Rates per 100,000 with 95% confidence intervals	p-value by group
Sex			0.795
Female	497	595.2 (544.1, 649.7)	
Male	456	584.7 (532.4, 640.8)	
Age group			<0.001
<5	61	553.3 (423.5, 710.2)	
5–19	184	532.4 (458.4, 614.9)	
20–59	326	425.5 (380.6, 474.1)	
≥60	382	972.4 (877.7, 1074.4)	
Ethnicity			<0.001
Māori	100	296.4 (241.2, 360.3)	
Pacific	17	284.0 (165.6, 454.4)	
Asian	29	580.2 (388.9, 832.3)	
European	802	716.8 (668.2, 768.0)*	
Other	5	146.1 (47.5, 340.6)	
Age group amongst hospitalized			<0.001
<5	1	9.1 (0.2, 50.5)	
5–19	1	2.9 (0.1, 16.1)	
20–59	7	9.1 (3.7, 18.8)	
≥60	31	78.9 (53.6, 112.0)*	
Serious outcomes			
Deceased	4		
Guillain-Barré syndrome	3		

*Rate in European ethnicity and hospitalized ≥60 years is significantly higher than other sub populations.

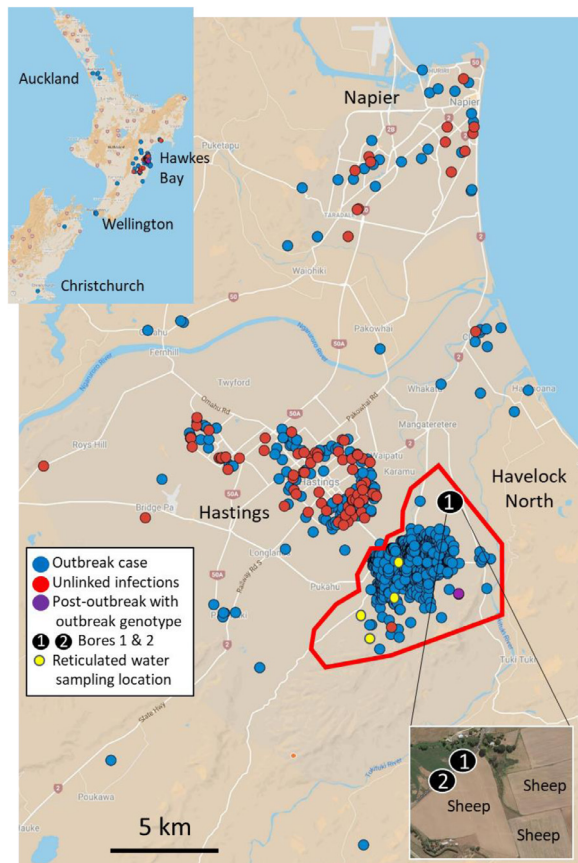


Fig. 2. Geographic distribution of outbreak cases, and unlinked infections with onset of illness of 7–24 August 2016. Also shown is the Havelock North contaminated reticulation area (red box), and locations of the reticulated network sampling points, bores and sheep paddocks.

(95% CI 6.0–8.0) for notified cases living in Havelock North. Applying this ratio to the 190 reported outbreak cases from outside Havelock North, would equate to an additional 1380 cases (95% CI 1250–1520). There was also a significant increase in campylobacte-

riosis notifications from the Hawke's Bay region with onsets between 7 and 24 August, who, when interviewed did not recall drinking Havelock North water (103 compared to 13 expected). Inclusion of these 90 cases in the outbreak, when multiplied by 7.3, would add another 650 (95% CI 590–720) cases to create a total of 7570 (95% CI 6850–8320) illnesses linked to the outbreak.

Microbiological investigation

Testing of four Havelock North reticulated water samples, and the bores sampled on 12 August found total coliforms of 30–110 MPN/100 mL and *E. coli* of 10–23 MPN/100 mL. All four reticulated water samples and one of the bore water samples were positive for *C. jejuni*. Following chlorination of the water supply on the 12 August, free available chlorine levels of 1 ppm were confirmed on 13 August. *C. jejuni* were isolated from 12 of 16 sheep fecal samples collected from paddocks adjacent to the bores.

Core genome MLST was undertaken using assembled genomes from 149 clinical, 11 water, and 19 sheep isolates (Table S1). The sequenced clinical isolates were from 88 confirmed cases, 14 unlinked infections from Hawke's Bay with onsets between 7 and 24 August, 2 infections with onsets prior to 7 August, and 43 infections with onsets after 24 August. Twelve core genome MLST genotypes were identified among the 88 confirmed case isolates, with the three most frequent being ST42-A (35 isolates), ST3610-A (25 isolates) and ST42-B (14 isolates) (Fig. 1B). At least two cases were infected with more than one genotype of *C. jejuni*. Four of the genotypes found in clinical cases were found in water isolates, three of them in sheep isolates, and one (ST42-A) in both water and sheep (Fig. 3). Within each of these cgMLST clusters, over 90% of the isolates had zero cgMLST differences. Of the 190 non-Havelock North outbreak cases (clinician confirmed cases who reported drinking Havelock North water), 62 were culture positive, and all 22 of those which were whole genome sequenced, were of an outbreak-associated genotype. Of the 23 cases from outside the Hawke's Bay, 20 were culture positive for *Campylobacter*, and two of these were whole genome sequenced. One of these was from Auckland with an outbreak-associated genotype of ST520-A, and the other from the Christchurch region with an outbreak-associated genotype of ST42-B. All of the 14 Hawke's Bay unlinked cases with onsets between 7 and 24 August that were

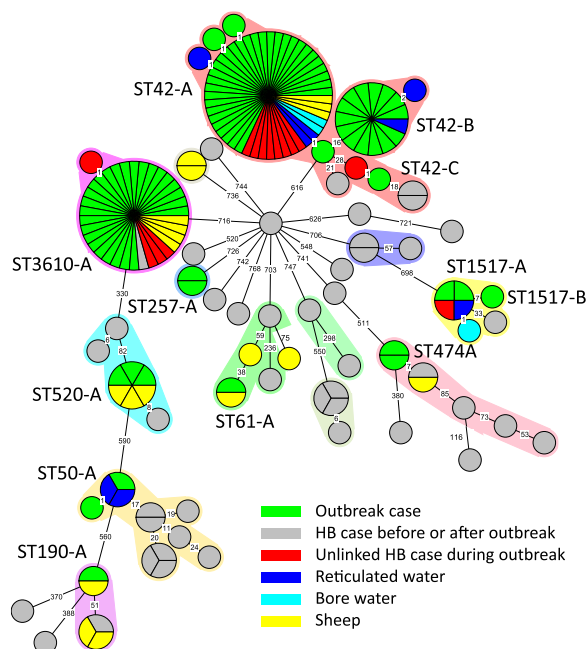


Fig. 3. Minimum spanning tree of core genome multi-locus sequence types (cgMLST) of isolates from outbreak cases, unlinked cases during the outbreak period, and reticulated water, bore water and sheep feces from paddocks adjacent to the drinking water source. Numbers on branches are the number of cgMLST differences between isolates, with isolates within the same circle indistinguishable when compared using the 1343 loci cgMLST scheme. Shading indicates that isolates with the same seven gene MLST type are labelled, with the letter after the outbreak indicating sub-clusters with fewer than three cgMLST differences.

genotyped clustered with isolates from outbreak cases, including the two most frequently observed cgMLST clusters. The 45 isolates from infections with onsets outside the outbreak period could be distinguished into at least 37 cgMLST genotypes, with only one of these genotypes (ST3610-A) indistinguishable from those found in outbreak cases. This was isolated from a two-year-old female from Havelock North with a symptom onset of 12 September 2016.

Discussion

We describe an exceptionally large *Campylobacter* waterborne outbreak that occurred following a heavy rainfall event which caused contamination of an untreated water supply. The most likely pathway of contamination was from sheep feces contaminating a stream which was hydrologically connected to the bore intake.¹⁰ This mechanism is supported by the high incidence of disease within the exposed population and identification of highly homologous genotypic clusters shared between isolates from affected humans, the contaminated water source, and sheep feces in nearby paddocks. The environmental events and systems failures that contributed to this outbreak occurring resulted in a comprehensive review of drinking water management in New Zealand and major reorganization of this sector.¹⁰

Reported rates of campylobacteriosis are typically higher in males, in those aged between 20 and 29, and children less than 5 years of age.¹¹ In this outbreak, reported rates did not show this pattern, perhaps reflecting the indiscriminate nature of drinking water exposure. Rates were disproportionately high among those aged over 60, which we attribute to underlying susceptibility in this age group¹², and the use of active case finding targeted to older residents.¹³

The ratio of cases estimated from the telephone survey ($n=5540$), to notified cases living in Havelock North ($n=763$)

of 7.3 (95% CI 6.6–8.0), is on the lower end of expected case-ascertainment ratio estimates. In the United Kingdom, one out of every 9.3 cases of campylobacteriosis is reported to national surveillance bodies,¹⁴ in the Netherlands this has been estimated at one out of 12,¹⁵ and in the USA one out of 30.3.¹⁶ It is possible that publicity around the event resulted in a higher proportion of people with diarrheal symptoms presenting to their doctors than usual.

While there have been larger documented waterborne outbreaks caused by protozoa,² the estimated total of between 6260 and 8320 cases is the largest ever reported outbreak caused by *Campylobacter*.³ Over a quarter lived outside the Havelock North area, emphasising the impact of events such as this on neighboring communities. We have included in this total, reported cases from the Hawke's Bay who were initially excluded from the outbreak because they did not recall drinking Havelock North water. Genotyping supported inclusion of these cases, most lived between 2 and 10 km from Havelock North and had an illness onset between 8 and 16 August, corresponding directly to the outbreak peak. Case interviews did not reveal any other common exposure. The drinking water exposure question may have been too specific as this may have excluded people who ate food prepared in Havelock North using the drinking water, visited a café, restaurant or private residence in Havelock North, washed their hands with Havelock North water, brushed their teeth with Havelock North water, or were otherwise exposed to the water. Secondary transmission is generally thought of as an uncommon transmission route for *Campylobacter*,¹⁷ and this study provides strong support for that finding, with only one clinical isolate, from a case with symptom onset after the 24 August (10 days post chlorination), having an outbreak-associated genotype.

Campylobacter infections have been previously reported to have an incubation period of between 1 and 10 days, with a peak after 3–4 days.¹⁸ In this paper the increase in notified infections, genotyping and analysis of visitors with a narrow water consumption period, suggest consumption of contaminated water from as early as 6 August. This timing correlates well with the heavy rainfall event beginning on 5 August. The water was contaminated through until the 13 August when chlorination of the system was achieved – a 7-day exposure period. With 99% of cases occurring within 10 days from the last date of water contamination, this timing supports the combined incubation periods previously reported.¹⁸

Mandatory laboratory notification of *Campylobacter* facilitated detection of the outbreak, allowing for prompt declaration of the 'boil water notice', initiation of water chlorination to prevent further spread of disease, and timely environmental sampling. The isolation and genotyping of *Campylobacter* from clinical, water and sheep specimens was invaluable in helping to identify the source of the outbreak, and confirmation of outbreak periods and cases. While there were at least twelve genotypes of *Campylobacter* among outbreak cases, 80% of the isolates were one of three genotypes (ST3610-A, ST42-A and ST42-B). With only one to three examples of each of the other genotypes, the isolation of indistinguishable genotypes from water or sheep feces for five of the other genotypes provides support for inclusion of these cases in the outbreak. The other four genotypes may relate to sources other than this outbreak (for example food or direct animal contact), or it is possible that additional testing of clinical, water or sheep isolates would have generated support for their epidemiological inclusion in the outbreak. Sequencing of only a few isolates could have generated a misleading picture of the outbreak, with different genotypes suggesting unrelated sources. In many waterborne outbreaks, recovery of pathogens from the water was unsuccessful probably because of the transient nature of contamination and delays in initiating testing. However, testing samples from the suspected contaminant sources (sheep in this outbreak, cattle in previous out-

breaks¹⁹), is a valid strategy, with strains potentially carried for months after an event.

The maturation of sequencing technologies makes the approaches used in this outbreak applicable to a broad range of large and small outbreaks. This situation is however threatened by culture-independent diagnostic testing.²⁰ Until methods are developed which integrate genotyping with these diagnostics, it is essential to continue to isolate and genotype causative organisms.

The whole genome sequencing demonstrated the value of this method in augmenting the epidemiologic investigation by discriminating between outbreak-linked and unlinked cases, confirming the suspected exposure period, identifying the likely source and mechanism of contamination, and defining the geographical scope of the outbreak. This investigation also showed the usefulness of supplementary epidemiological investigations, notably cross-sectional surveys of the exposed population during the outbreak, to rapidly assess the outbreak magnitude, its population impact and to support early response.

A campylobacteriosis outbreak of this magnitude casts a spotlight on the global issue of groundwater security in the setting of heavy rainfall,²¹ the risk of waterborne outbreaks in non-chlorinated drinking water supplies,^{22,23} risks to water sources from animal feces,^{19,24} and the importance of monitoring for contamination following heavy rainfall events. Climate disruption is likely to increase the frequency of such events²⁵. It therefore becomes paramount to establish safeguards to prevent contamination of groundwater supplies and to promptly detect breaches in order to protect populations, particularly in regions where intensive agriculture results in high faecal contamination of water catchments. Contamination of groundwater from livestock faeces was also identified as a likely source in the Walkerton, Canada outbreak in 2000.¹⁹ A governmental inquiry into this Havelock North outbreak has recommended a complete overhaul of drinking water management in New Zealand. One immediate change was that all drinking water supplies, including groundwater, should be treated and that residual disinfection should be required to mitigate the risk of treated water contamination within the supply reticulation.¹⁰

Contributors

TW, SP, JS, GM, TH, CH, RE, and NJ undertook epidemiological investigations; AB, MW, PS, BR, SL, AC, LR, AM and PB undertook isolation of *Campylobacter* and whole genome sequencing; with JZ, DW and BG doing bioinformatic analysis; TW designed and undertook statistical analyses; NJ, MB, BG wrote the original funding application; TW, BG, SP, JS, DH, NF, AJ, MB, and NJ undertook the writing and revising of this paper; BG created the figures. All authors commented on the draft manuscript and contributed to the final version.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.jinf.2020.06.065.

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