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An outbreak of norovirus GI-6 following a wedding in North West England

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Summary

In August 2015 a gastroenteritis outbreak occurred following a wedding. An outbreak investigation was undertaken and a cohort study was conducted using an online survey. Of 140 guests, 134 received the survey and 113 responded (84.3% response rate). 70 respondents met the case definition of vomiting and/or diarrhoea within 72 hours of the wedding (61.9% attack rate). Fifteen exposures were associated with illness; on stratification, all were confounded by the ham hock starter. Multivariable analysis showed a significant association with exposure to ham hock (RR 6.62, 95% CI: 2.19-20.03). Eight guests and two catering staff submitted stool samples. All tested positive for norovirus GI-6, including a food-handler who had vomiting less than 48 hours before the wedding. A single genotype was detected amongst all samples, suggesting a single source of contamination. The transmission pattern suggested point-source exposure. The most plausible cause of the outbreak was transmission from an infected food-handler via contaminated food. This highlights the importance of appropriate exclusions for symptomatic food-handlers. Additionally, the food-handler's stool sample was submitted 7 days after symptom resolution. The potential for extended viral excretion, and the extremely low infective dose of norovirus, may mean that current exclusion guidelines are not of sufficient duration.

Introduction

Norovirus is a highly infectious viral pathogen of the *Caliciviridae* family that is a common cause of gastroenteritis in adults and children [1]. Norovirus was found to be the most common cause of infectious intestinal disease in the UK during the IID2 longitudinal study [2]. Additionally, using 2008-2009 prices, norovirus was estimated to have a total annual cost to patients and the health service of £81 million (95% CI: £63 million - £106 million), which, based on the same study, was a larger economic burden than that of campylobacter and rotavirus combined [3].

Infected individuals excrete the virus in faeces and vomit, and continue to do so for several days, before, during and after their symptoms. Asymptomatic infection also occurs, and these individuals can also shed the virus, posing a particular challenge to infection control [4] [5] [6]. Humans are the only known reservoir for human norovirus infections, but contamination of food, water and the environment is possible, so indirect transmission of norovirus also occurs [4] [7] [8] [9].

This paper describes an investigation into a food-associated norovirus outbreak linked to a wedding in the North West. In August 2015, Public Health England (PHE) were contacted about a wedding, with reports that over 50 of the 140 guests were experiencing symptoms of illness including diarrhoea, vomiting and headache. An outbreak was declared and an Outbreak Control Team (OCT) was convened to undertake an outbreak investigation, identify the source of the outbreak and ensure appropriate control measures were implemented to prevent further transmission.

Methods

Epidemiological Investigations

Cases were defined as any person who attended the wedding and experienced vomiting and/or diarrhoea with an onset date in the 72 hours following the wedding.

A retrospective cohort study was carried out to support the outbreak investigation, using a list of food and drink items provided by the caterers. A web-based survey was sent to people who attended the wedding, and was first emailed to the bride's family for circulation six days after the wedding. Respondents could complete the survey on behalf of other guests, such as young children, and were asked to indicate if they had done so. Staff who worked at the wedding were excluded from the cohort as their exposures were believed to be systematically different from guests.

Demographics and exposure amongst cases and non-cases were described and compared. Univariable and multivariable analysis was undertaken. Poisson regression with robust error variance was used to quantify associations between exposures and illness (Risk Ratios (RR) with 95% Confidence Interval (95% CI)).

For each item with a p value <0.05 on univariable analysis, univariable regression was conducted to explore if any of the associations showed a dose-response relationship. For canapés, respondents were asked to select from none, 1-2 or 3+. For other food items, respondents were asked to select from none, a taste or a portion. Exposure to drink items and the chocolate truffles was not quantified in the survey questions.

The multivariable analysis was undertaken using forward stepwise modelling. Only those items which significantly improved model parsimony, as measured using the Akaike information criterion (AIC), were included in the final model.

All analyses were undertaken using Stata v12.1.

Microbiological Investigations

Stool specimens were submitted by two catering staff and eight wedding guests. The eight wedding guests came from four different households. The 10 samples were collected between 5 and 13 days after the wedding. All 10 samples were tested for bacteria and enteric viruses. Environmental swabs were also taken at the caterer's premises and the wedding venue, and were tested for hygiene

indicator organisms (including enterobacteriaceae, E.coli and coagulase positive Staphylococci) and norovirus.

For faecal specimens, total nucleic acid was obtained using QIAxtractor automated nucleic acid extraction platform (QIAGEN). For environmental specimens, total nucleic acid was extracted using a GTC–silica method performed as previously described [10]. From total nucleic acid, RNA was converted to cDNA in a random-primed reverse transcription reaction and norovirus detected using real-time PCR methods as previously described [11].

Noroviruses were genotyped through nucleic acid sequence analysis of a region encoding the S-domain of the capsid (region C) [12] [13]. Sequence analysis was performed using Bionumerics v6.1 (Applied Maths, Kortijk,Belgium).

Environmental Investigations

Environmental Health Officers (EHOs) conducted environmental inspections of the caterer's premises and the wedding venue, along with review of the caterer's Food Safety Management System and discussions with staff from both companies. No food samples were submitted as none were remaining from the event.

Results

Epidemiological Investigations

The survey was sent to 134 of 140 wedding guests. In total, 113 (83.4%) individuals responded to the survey, of whom 60 (53.1%) were male. Age was reported by 89 respondents, with an average age of 39, ranging from 4-88.

Of the survey respondents, 76 (67.3%) reported illness. However, one gave an onset date greater than 72 hours after the event and 5 reported illness but did not have symptoms of vomiting or diarrhoea, so were not included as cases. Therefore, of the 113 respondents, 70 (61.9%) met the case definition.

Of the 70 cases, 50 (71.4%) experienced vomiting, and 64 (91.4%) experienced diarrhoea. Nausea was reported by 65 (92.9%) and abdominal pain was reported by 51 (72.9%) people. Over half of cases also experienced fever (54.3%), and half reported headache (50.0%), with a small number (7.1%) reporting bloody diarrhoea. The most commonly reported symptom duration was 2 days (48.6%), with duration ranging from less than one day to five or more.

There were no significant differences between the cases and non-cases by age or gender. Individuals who attended the event during the whole day, which included the canape reception and wedding breakfast as well as the evening buffet, had an estimated relative risk (RR) of 4.42 (95% CI 1.49-11.99) of becoming a case when compared with those who attended only the evening buffet.

The analytical study identified 15 items where there was strong evidence of association between exposure and being a case, as shown in Table 1. At a univariable level, the highest RRs were for the ham hock starter (RR 7.63, 95% CI 2.60-22.32), the meat sausages served to adults (RR 6.91, 95% CI 2.37-20.15), and the mashed potato (RR 5.35, 95% CI 2.15-13.31) served as an accompaniment to all of the main courses. Of the 70 cases, 67 (95.7%) ate the ham hock and meat sausages and 65 (92.8%) ate the mashed potatoes.

For the dose-response analysis the canapés either showed a lower effect at a higher dose, or did not have strong evidence for a dose-response relationship. The ham hock starter, meat sausages, and leeks all showed strong dose-response relationships. The mashed potato, peas and lemon meringue all showed some dose-response. The bread basket showed no evidence of a dose-response relationship.

The mashed potato, peas, leeks, bread basket, lemon meringue, chocolate truffles and all of the canapés were confounded by the ham hock. The champagne was also confounded by the ham hock with some evidence of effect modification. The mineral water showed possible confounding.

Due to strong collinearity between the ham hock and the meat sausages it was not possible to assess confounding. Consequently, for subsequent modelling, the ham hock was used and the meat sausages were excluded in view of the higher univariable risk associated with the ham hock.

The final multivariable model is shown in Table 2; inclusion of the leeks, the chicken skewers, the cheese risotto, and the mineral water significantly improved the model. When adjusted for these items, there was still a strongly significant relationship between becoming a case and having been exposed to the ham hock (adjusted RR 6.62, 95% CI: 2.19-20.03).

In addition to the quantitative analysis, qualitative comments were also received via the survey. Of 45 text responses, ten people (22.2%) mentioned the ham hock, with comments including concerns about the serving temperature, the smell, the taste, the texture and the meat being undercooked. One person (2.2%) commented that the lemon meringue was sour, but no other comments were received about specific food items.

Microbiological Investigations

Stool Samples

The 8 specimens submitted from wedding guests for testing were negative for any pathogenic bacteria, however, genogroup I norovirus was detected in all eight samples.

Two staff members who catered the event submitted specimens and genogroup I norovirus was detected in both. One of these individuals reported symptoms prior to the event, and the other reported symptoms that began on the day of the event. A third symptomatic staff member did not submit a specimen.

Environmental Samples

Of the 17 swabs taken from the caterers' premises, 7 were tested for hygiene indicator organisms and 10 were tested for norovirus. A hygiene indicator sample from the chiller handle tested positive for Enterobacteriaceae.

Twenty swabs were taken from the venue and tested for norovirus. A genogroup II norovirus was detected in one swab from the ladies' toilet and one from the chiller handle. The venue had not been open to the public between the wedding and the sampling. However, venue staff had access to the chiller, which they used to store milk, and the public toilets in the intervening period.

Additional Testing

All of the positive samples were sent to the national reference laboratory for further typing. Further analysis was conducted for all 10 stool samples (8 guests and 2 staff members). The genogroup I virus was characterised as genotype 6 (GI-6); all sequences were identical. No further analysis was possible on the environmental swabs.

Environmental Investigations

<u>Wedding Venue</u>

An EHO visit did not identify any areas of concern. The venue staff were not involved with catering and none reported being unwell before, during or after the wedding. The norovirus identified on environmental swabs was different to that involved in the outbreak; however, in view of its presence the EHO provided reminders of handwashing advice to venue staff.

<u>Caterer</u>

The investigation identified a number of areas of concern. The chef who provided catering on the wedding day had vomiting which had resolved less than 48 hours earlier, and a GI-6 norovirus was detected in a faecal specimen submitted 5 days after the wedding. Two staff were ill at the wedding

and were quarantined at the venue until the end of the event; one submitted a specimen in which GI-6 norovirus was detected.

The ham hock starter was cooked two days prior to the wedding by a chef who did not attend the event and did not report illness. It was transported to the venue in moulds on the wedding day, when it was plated and garnished, without further cooking, by the chef who subsequently tested positive for norovirus. There were no records of control checks to assess storage, transport, hot holding or service temperatures. A hand wash basin at the caterer's premises was next to a raw meat preparation area.

As a result of the findings of the outbreak investigation, a warning letter was issued by the local authority environmental health team.

Discussion

Our investigation concluded that this outbreak of norovirus gastroenteritis was associated with a ham hock dish that had been prepared by a food-handler who had vomiting less than 48 hours before the wedding. A GI-6 norovirus was detected in the faeces of all cases and food handlers tested. The food-handler who had vomiting less than 48 hours before the wedding submitted a specimen five days after the wedding in which a GI-6 virus was detected, having had no further symptoms since the vomiting resolved.

The epidemiological analysis indicated a strong association between the ham hock and becoming a case. The collinearity between the ham hock and the meat sausages meant that the sausages were excluded from further analysis, and this is a limitation of the study. It was also noted that three of the cases did not report consuming the ham hock. The lack of any food samples for testing also meant that it was not possible to prove that any items were contaminated with norovirus. However,

179 the epidemiological analysis, including the qualitative feedback in the survey, alongside the findings
180 of the environmental investigation, lead to the conclusion that the ham hock was the most likely
181 vehicle of transmission. Furthermore, the positive stool sample in a food-handler who was
182 symptomatic prior to the wedding provided a plausible source of contamination of the food.

183 Although GI-6 norovirus strains are detected in the UK each year, over the last 10 years they have
184 only accounted for 1.0-1.5% of all norovirus strains identified (DJA personal communication). A
185 paper on diversity of norovirus in the North of England included two outbreaks which were
186 designated as GI-6 [13]. Publications have also reported outbreaks linked with GI-6 in Hesse,
187 Germany [14], the USA [15] and Australia [16].

188 The Centers for Disease Control and Prevention in the USA, reported in 2013 that there had been an
189 increase in the proportion of GI-6 outbreaks in recent years, with 7.7% of 2012 outbreaks being due
190 to GI-6 compared with 1.4% in 2010. Additionally, it was found that GI-6 outbreaks were more likely
191 to be associated with food, and had summer seasonality, when compared with non-GI-6 outbreaks
192 [17]. This correlates with the timing and mode of transmission of the outbreak investigations
193 detailed in this paper.

194 The IID2 study has identified that the burden of norovirus in the UK is likely to be much higher than
195 that identified by national surveillance. It is estimated that for every norovirus case identified
196 through surveillance, there were 12.7 GP consultations (95% CI 8.8-18.3), and 288 cases in the
197 community (95% CI 239-346) [2]. Furthermore, as GI-6 outbreaks have been shown to be less likely
198 to occur in healthcare settings than non-GI-6 outbreaks [17], it may be that there is an element of
199 underreporting when compared with the strains that commonly occur in healthcare settings, due to
200 a decreased likelihood of symptom reporting and sample submission.

201 The food-associated transmission in this outbreak was associated with contamination by an infected
202 food handler, who was not appropriately excluded for 48 hours following the resolution of

symptoms. There have been multiple papers outlining the role of post-symptomatic food handlers in the transmission of norovirus and subsequent outbreaks [8, 6, 7]. This outbreak further highlights the importance of appropriate exclusions for symptomatic food handlers.

Whilst the food handler returned to work less than 48 hours after symptom resolution, the positive stool sample 7 days after reported resolution of symptoms also reinforces the potential for extended viral excretion in those infected. It has been demonstrated in that post-symptomatic viral shedding of norovirus may last longer than the typical 48 hour exclusion period [18, 19]. Particularly in view of the low infective dose of norovirus, this extended, post-symptomatic excretion, could well be an ongoing risk for transmission. Therefore, as previously highlighted, in food handlers with gastroenteritis symptoms, there should be further consideration of whether the existing exclusion guidelines are sufficient [5].

Conclusions

This study found that this norovirus outbreak was food-associated transmission of norovirus (GI-6), associated with inappropriate exclusion of an infected food-handler. The summer seasonality and mode of transmission are in keeping with broader trends related to GI-6 in the US. The extended post-symptomatic viral excretion in the food handler highlights the need for further consideration of appropriate duration of exclusion in this group.

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Conflict of Interest

None

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Table 1 – Univariable Analysis

Exposure	Exposed			Unexposed			Risk Ratio [95% CI]	p Value
	Total	Case	AR%	Total	Case	AR%		
Ham Hock	82	67	81.7	28	3	10.7	7.63 [2.60-22.32]	<0.0001
Meat Sausages	84	67	79.8	26	3	11.5	6.91 [2.37-20.15]	<0.0001
Mashed Potato	82	65	79.3	27	4	14.8	5.35 [2.15-13.31]	<0.0001
Lemon Meringue	80	63	78.8	31	7	22.6	3.49 [1.80-6.76]	<0.0001
Peas	69	56	81.2	38	11	29.0	2.80 [1.68-4.67]	<0.0001
Leeks	59	49	83.1	44	15	34.1	2.44 [1.59-3.73]	<0.0001
Chicken Skewers	59	48	81.4	47	19	40.4	2.01 [1.39-2.91]	<0.0001
Champagne	67	53	79.1	40	16	40.0	1.98 [1.33-2.95]	<0.0001
Bread Basket	66	51	77.3	41	16	39.0	1.98 [1.32-2.97]	<0.0001
Cheese Risotto Balls	57	45	79.0	47	19	40.4	1.95 [1.35-2.83]	<0.0001
Tandoori Chicken	52	42	80.7	55	25	55.5	1.78 [1.29-2.44]	<0.0001
Spiced Lamb	54	41	75.9	54	27	50.0	1.52 [1.12-2.06]	0.009
Mushroom Risotto	42	33	78.6	63	33	52.4	1.50 [1.13-1.99]	0.008
Mineral Water	39	29	74.4	60	30	50.0	1.49 [1.09-2.03]	0.021
Chocolate Truffles	42	32	76.2	68	38	55.9	1.36 [1.04-1.79]	0.041

Table 2 – Multivariable Model

Exposure	Adjusted Risk Ratio [95% CI]	P
Ham Hock	6.62 [2.19 - 20.03]	0.001
Leeks	1.15 [0.84 - 1.57]	0.397
Chicken Skewers	0.91 [0.69 - 1.20]	0.912
Cheese Risotto Balls	1.01 [0.77 - 1.33]	0.943
Mineral Water	1.27 [0.98 – 1.64]	0.071

Figure 1: Epidemic curve showing onset date of illness in persons associated with the wedding, August, 2015

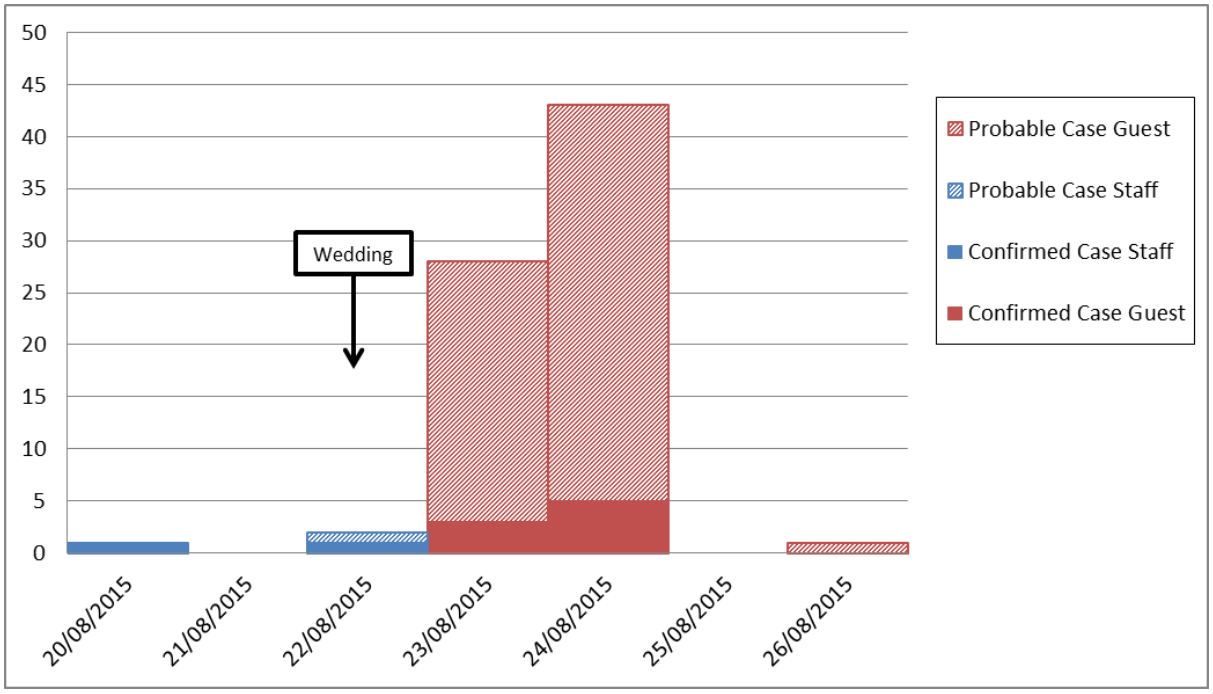

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Figure 2: Phylogenetic tree derived from 8 norovirus partial capsid (ORF2) sequences from 8 specimens collected from symptomatic guests. The tree was constructed using the Maximum Likelihood method in MEGA6 [20]. Branch length represents number of substitutions per site. Specimens from cases are marked with blue triangles; reference sequences obtained from GenBank are marked with grey squares. Numbers on branches show bootstrap values >80% (1000 replicates).