

ETEC outbreak during a conference in Oslo

April 2016

Collaborating partners

- **Norwegian Institute of Public Health**
- Hinta Meijerink, Heidi Lange, Katrine Borgen, Nils O. Hermansen, Lin T. Brandal, Solveig Jore, Karin Nygård, Line Vold
- **Norwegian Food Safety Authority, department of Oslo, Asker and Bærum**
Kari Dommarsnes, Solveig Eriksrud, Marit Kolle
- **Infection disease control in Oslo, Municipal doctor in Oslo**
Tore Steen

Contact information

utbrudd@fhi.no

Table of Contents

Sammendrag	2
Summary	2
Background	3
Overview of the event	3
Methods.....	3
Epidemiological investigation	3
Microbiological investigation.....	4
Environmental investigation	4
Results.....	4
Epidemiological investigation	4
Microbiological investigation.....	6
Environmental investigation	7
Discussion.....	7
References	8

Sammendrag

Mandag 14. mars mottok Folkehelseinstituttet varsel om 40 tilfeller av gastroenteritt blant 590 deltakere ved en konferanse avholdt 10. og 11. mars i Oslo. I nært samarbeid med Mattilsynet, kommunelegen og konferansearrangør ble en utbruddsetterforskning igangsatt. 15. mars sendte vi ut et kort online spørreskjema til alle deltagere på konferansen. Det inneholdt blant annet spørsmål om alle måltider servert under konferansen. Et tilfelle ble definert som en person som deltok på konferansen, rapporterte diaré eller to av følgende symptomer: oppkast, kvalme, magesmerter, hodepine, feber med varighet på minst 2 dager. Totalt 453 av de 590 deltakerne (77%) besvarte spørreskjemaet og 110 (25%) oppfylte kasusdefinisjonen. Symptomene rapportert blant tilfellene inkluderte diaré (90%), magesmerter (88%), kvalme (65%), hodepine (45%), leddsmerte (27%), feber (27%) og oppkast (12%). Symptomstart ble rapportert fra kl. 13 på torsdag (10. mars) til kl. 11 på mandag (14. mars). De fleste tilfellene (43%) var fortsatt syke da de fylte ut spørreskjemaet. Basert på epidemiologiske undersøkelser ble torsk servert til lunsj på torsdag 10. mars identifisert som mest sannsynlig smittetilstand (RR = 4,1; 95% CI: 2,8 - 6,1). Torsken var en av få varmretter og den ble pyntet med frisk gressløk etter varmebehandling. Totalt ble avføringsprøver fra 18 pasienter undersøkt; 12 prøver var positive for ETEC og 10 var positive for EPEC. Miljøundersøkelser avdekket ingen alvorlige brudd på kjøkkenhygiene ved konferansesenteret. Gressløk fra samme batch ble testet og var negativ for ETEC, EPEC og *E.coli*. På bakgrunn av tydelige epidemiologiske funn, identifisering av ETEC i alle avføringsprøver testet, gunstige oppformeringsforhold for *E.coli* på den varme torsken, og tidligere ETEC utbrudd relatert til gressløk konkluderte vi med at gressløk kontaminert med ETEC var den mest sannsynlige årsaken til dette utbruddet. Mattilsynet frarådet bruk av gressløk fra samme parti (160kg) for matlaging og understreket rådet om varmebehandling av friske urter før bruk, evt. å tilsette friske urter rett før konsum.

Summary

On Monday 14 March, the Norwegian Institute of Public Health (NIPH) received a report of 40 cases of gastroenteritis among 590 participants of a conference on 10 and 11 March. In close contact with the regional office of the Norwegian Food Safety Authorities (NFSA), the municipal health officer and the conference organisers, we initiated an outbreak investigation. On 15 March, we send out a short self-administered online questionnaire including all food items served during the conference. A case was defined as an individual who attended the conference, reported diarrhoea or two of the following symptoms: vomiting, nausea, stomach ache, headache, fever with duration of at least 2 days. In total, 453 of the 590 participants (77%) completed the questionnaire and 110 (25%) fitted the case definition. Symptoms reported among the cases included diarrhoea (90%), stomach pain (88%), nausea (65%), headache (45%), joint pain (27%), fever (27%) and vomiting (12%). Onset of symptoms was reported from 1pm on Thursday (10 March) to 11am on Monday (14 March). Most of the cases reported still to be ill when they filled out the questionnaire (43%). Based on univariable and multivariable analyses cod served for lunch on Thursday 10 March was associated with illness (RR=4.1; 95%CI: 2.8-6.1). The cod was one of the few warm dishes served and it was garnished with fresh chives subsequent to heat treatment, providing favourable conditions for the growth of bacteria. In total, we received samples from 18 patients; 12 samples were positive for ETEC and 10 samples were positive for EPEC. Environmental investigation did not identify any serious hygiene relapses in the kitchen. Samples of the chives tested negative for ETEC, EPEC and *E.coli*. Based on strong epidemiological evidence, identification of ETEC in all stool samples, previous ETEC outbreaks related chives and biological plausibility, we conclude that chives contaminated with ETEC were the most likely source of this outbreak. The NFSA recommended not using chives from the same batch (160kg) for food preparation. In addition, we reinforced the advice on heat treatment of fresh herbs before usage or adding fresh herbs right before consumption.

Background

Overview of the event

On Monday 14 March, the organisers of a conference in Oslo contacted the local department of the Norwegian Food Safety Authorities (NFSA) and the municipal doctor in Oslo and informed them that a number of participants of the conference fell ill with gastrointestinal symptoms. On Tuesday 15 March, the NFSA reported 60 cases of gastroenteritis among the 590 participants of the conference to the Norwegian Institute of Public Health (NIPH).

The conference took place on 10 and 11 March 2016 at a hotel in Oslo. Participants of the conference were provided snacks and lunch on both days. In addition, some participants joined an organised dinner on Thursday 10 March. An outbreak investigation was initiated by the Oslo department of the NFSA in collaboration with NIPH on Tuesday 15 March 2016. The objective of the investigation was to ascertain the extent of the outbreak and identify the source in order to implement control measures, prevent further spread and give recommendations.

Methods

Epidemiological investigation

We conducted a retrospective cohort study including all the 590 participants of the conference that took place at a hotel in Oslo on 10 and 11 March 2016.

Case definition

A case was defined as a person who attended the conference on 10 and/or 11 March 2016, reported having onset of diarrhoea or two of the following symptoms: vomiting, nausea, stomach ache, headache, fever between Thursday 10 and Monday 14 March, with a duration of at least 2 days (or “still ill” when answering the questionnaire).

Data collection

To investigate potential food exposures we developed a short self-administered questionnaire in the web-based tool Questback. The questionnaire included demographic factors, information on gastrointestinal illness (before, during and after the conference) and consumption of all food items served during the conference on 10 and 11 March 2016. The list of food items and drinks included were based on the menu obtained from the hotel. The questionnaire was piloted internally at the NIPH prior to dissemination. NIPH sent the questionnaire to the conference organisers, who distributed it to all participants via email on 16 March 2016. Data collection was closed by 1 April 2016. In the questionnaire, the exposure to food and drink items was categorized as follows: ate/drank item; probably ate/drank item; probably did not eat/drink item; did not eat/drink item.

Data analysis

Data was extracted from Questback into Excel and thereafter exported to and cleaned in Stata 14. Descriptive and univariable analyses were performed in Stata 14. We calculated the number of people exposed to the various food items, number of ill people among the exposed and unexposed and attack rates (AR) for all food items. Exposure was defined as reporting “ate” or “probably ate” with regards to that specific item. Risk ratios (RRs) were calculated for all exposures. We analysed the association of each food item and illness one by one (univariable analysis). In addition, we performed multivariable analyses on items served at the same time during the conference with an association with illness in the univariable analyses ($p < 0.200$).

Microbiological investigation

Patient samples

All symptomatic individuals were encouraged to see a medical doctor and deliver a stool sample. All stool samples in Norway are routinely analysed for *Salmonella*, *Yersinia*, *Shigella* and *Campylobacter*. For other pathogens, samples are tested on request. However, in some of the larger medical microbiology laboratories all faecal samples are screened for a large diversity of pathogens including also *E.coli*, norovirus and adenovirus.

Food samples

There were no leftovers to be sampled by the regional NFSA. As soon as chives were suspected from the preliminary epidemiological analyses, one (unopened) package of chives from the same batch that was used during the preparation of the conference lunch on Thursday was sampled and sent to Veterinærinstituttet. They tested for ETEC, EPEC and total number of *E.coli*.

Environmental investigation

The hotel was contacted and visited by NFSA for interviews on food preparation and inspection of the kitchen.

Results

Epidemiological investigation

Description of cases

The questionnaire was completed by 453 out of 590 registered conference participants, resulting in a response proportion of 77%. Five people did not attend the conference and were excluded from further analyses. We identified 109 cases, corresponding to an attack rate of 24.0%. The first case reported onset of symptom at 1pm on Thursday 10 June and the last case at 11am on Monday 14 March (Figure 1).

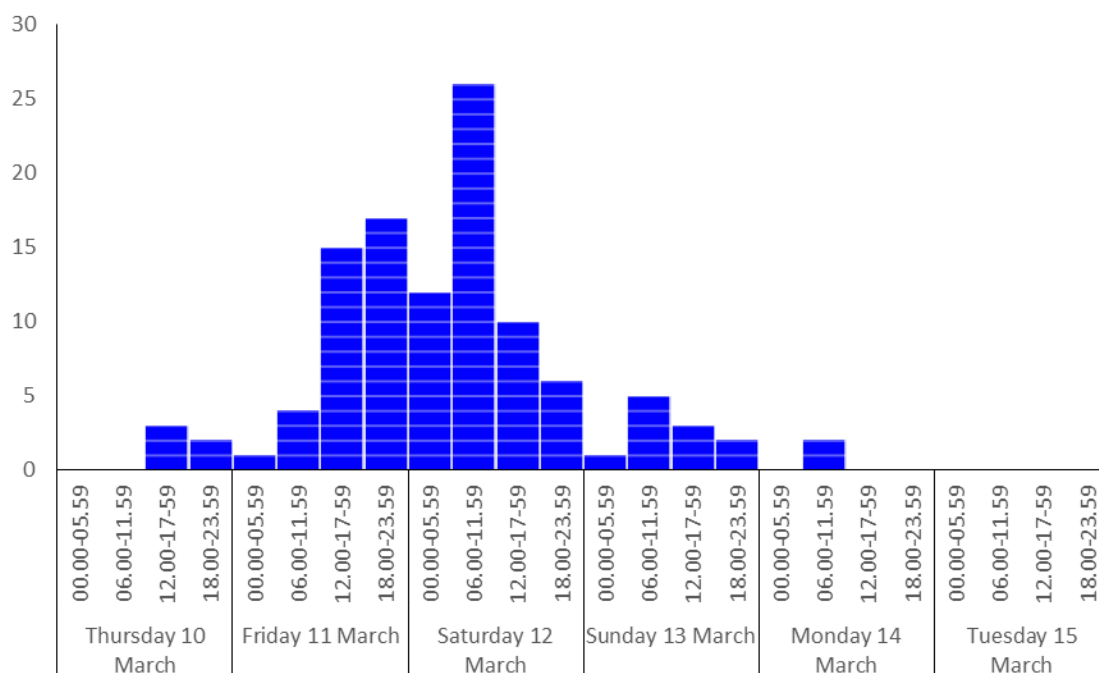


Figure 1. Distribution of individuals by onset of gastroenteritis symptoms among participants of a conference in Oslo, 10-11 March 2016.

The most common symptoms were diarrhoea, stomach pain and nausea (Table 1). The majority of cases were still ill when they filled out the questionnaire (42%), 15% were ill two days, 16% three days, 12% four days and 16% five or more days.

Table 1. Reported symptoms among participants of among participants of a conference in Oslo, 10-11 March 2016 (n=109).

Symptom	n	%
Diarrhoea	98	89.9
Stomach pain	96	88.1
Nausea	70	64.2
Headache	49	45.0
Joint pain	30	27.5
Fever	30	27.5
Vomiting	13	11.9

Analytical epidemiology

Based on reported onset of disease, Thursday 10 March was the most likely day of exposure and we therefore focussed our analyses on food served on this day. In univariable analyses, the cod and mixed salad were associated with illness (Table 2).

Table 2. Attack rates and risk ratios by food item among participants of a conference in Oslo, 10-11 March 2016 (n=109).

Lunch Thursday	Exposed			Unexposed			RR	95% CI	p	% cases exposed
	Total	Case	AR%	Total	Case	AR%				
Cod with chives	171	78	45.6	262	29	11.1	4.12	2.82-6.03	<0.001	72.9
Mixed salad	224	71	31.7	209	36	17.2	1.84	1.29-2.62	<0.001	66.4
Noodle salad	190	55	29.0	243	52	21.4	1.35	0.97-1.88	0.071	51.4
Strawberries	137	41	29.9	296	66	22.3	1.34	0.96-1.87	0.087	38.3
Watermelon	188	54	28.7	245	53	21.6	1.33	0.96-1.84	0.090	50.5
Turkey wrap	282	75	26.6	151	32	21.2	1.25	0.87-1.81	0.214	70.1
Cream	62	18	29.0	371	89	24.0	1.21	0.79-1.86	0.394	16.8
Tomato	163	43	26.4	270	64	23.7	1.11	0.80-1.55	0.532	40.2
Heart salad	191	50	26.2	242	57	23.6	1.11	0.80-1.54	0.530	46.7
Scones	148	39	26.4	285	68	23.9	1.10	0.79-1.55	0.569	36.4
Cheesecake	304	77	25.3	129	30	23.3	1.09	0.75-1.57	0.647	72.0
Jam	91	24	26.4	342	83	24.3	1.09	0.74-1.61	0.679	22.4
Honey melon	218	56	25.7	215	51	23.7	1.08	0.78-1.51	0.635	52.3
Muffins	183	46	25.1	250	61	24.4	1.03	0.74-1.44	0.861	43.0
Grapes	231	57	24.7	202	50	24.8	1.00	0.72-1.39	0.985	53.3
Blueberries	122	30	24.6	311	77	24.8	0.99	0.69-1.43	0.971	28.0
Tomato bread	285	69	24.2	148	38	25.7	0.94	0.67-1.33	0.737	64.5
Apple salad	104	24	23.1	329	83	25.2	0.91	0.61-1.36	0.658	22.4
Ketchup	84	18	21.4	349	89	25.5	0.84	0.54-1.31	0.437	16.8
Hamburger	172	38	22.1	261	69	26.4	0.84	0.59-1.18	0.305	35.5
Orange	152	33	21.7	281	74	26.3	0.82	0.58-1.18	0.287	30.8
Onion	112	20	17.9	321	87	27.1	0.66	0.43-1.02	0.051	18.7

For multivariable analyses we took into account all variables with an association with disease ($p < 0.200$) in the univariable analyses, i.e. cod with chives, mixed salad, noodle salad, strawberries and watermelon (Table 3). Cod was still strongly associated with illness (RR=3.88, 95%CI: 2.64-5.69), after adjustment for other possible exposures. Looking at proportion of cases exposed, 73% of cases could be explained by the cod. In total 93% (n=101) of all cases could be explained by the dishes that contained chives (cod and turkey wrap).

Table 3. Multivariable analyses of exposures associated with illness among participants of a conference in Oslo, 10-11 March 2016 (n=109).

Exposure	Risk Ratio	95%CI	p-value
Cod with chives	3.88	2.64 – 5.69	<0.001
Mixed salad	1.48	1.06 – 2.07	0.022
Noodle salad	0.96	0.72 – 1.29	0.808
Strawberries	1.22	0.91 – 1.64	0.187
Watermelon	1.16	0.86 – 1.57	0.323

Microbiological investigation

Patients

We received information about samples from 18 patients; 12 samples were positive for ETEC and 10 samples were positive for EPEC. The National Reference Laboratory at NIPH received samples from nine patients and ETEC was confirmed in six of them (Table 4). All except two ETEC isolates, showed different MLVA- and virulence- profiles. Two patients were infected with enteroaggregative *E. coli* (*aggR* positive), whereas the last patient carried enteropathogenic *E. coli* (*eae* positive).

Table 4. Results from the microbiological investigation of stool samples from gastroenteritis cases among participants of a conference in Oslo, 10-11 March 2016

Patient	ETEC	Virulence	Serotype	MLVA- profile
1	pos	<i>ST1b</i>	O?	5-1-0-8-3-1-1-16-0-0
2	pos	<i>ST1b+LT1</i>	O6	5-0-0-8-3-11-1-35-0-0
3	pos	<i>ST1a</i>	O?	6-1-0-8-3-3-1-6-0-0
4	pos	<i>LT1</i>	O?	7-3-0-8-3-13-1-6-6-0
	pos	<i>LT1</i>	O?	6-3-0-8-3-11-1-6-6-0
	neg	neg	O?	7-3-0-8-3-13-1-79-0-0
	neg	neg	O?	7-3-0-8-3-13-1-79-3-0
5	neg	<i>eae</i>	O?	5-18-0-8-3-3-1-6-6-0
	neg	neg	O?	7-3-0-8-3-7-1-79-0-0
6	neg	<i>aggR</i>	O?	5-3-5-8-4-3-1-6-7-0
7	pos	<i>ST1b+LT1</i>	O6	5-0-0-8-3-11-1-35-0-0
	neg	neg	O?	5-3-0-8-3-11-1-84-0-0
8	pos	<i>ST1a</i>	O?	6-1-0-8-3-3-1-6-0-0
9	neg	<i>aggR</i>	O?	6-3-0-8-3-8-1-6-11-0
	neg	neg	O?	5-0-9-8-3-8-1-0-0-0

Food

The suspected chives that were used with the cod served for lunch tested negative for ETEC and EPEC, and the total number of *E. coli* was below the detection limit (<10cfu/g).

Environmental investigation

The NFSA conducted an inspection of on Monday 14 March 2016. Environmental investigation did not identify any serious hygiene relapses in the kitchen. None of the kitchen staff had any gastrointestinal symptoms prior, during or after the event that could be linked to the outbreak. Based on the epidemiological analyses, the cod served topped with freshly cut chives were the suspected source of the outbreak and NFSA collected left-over chives from the hotel. These chives (imported from Kenya) were sent to the Veterinary institute in Oslo on 18 March 2016 for further microbiological analyses. When gathering more information about food preparation, we learned that the cod was one of the few warm dishes served. The chives were added after preparation of the cod and the cod (with chives) was placed in a buffet. The temperature and time before consumption of the cod could have provided favourable conditions for the growth of bacteria. The chives were also used in the turkey wrap, which was served cold.

Discussion

We have strong epidemiological evidence and biological plausibility that chives contaminated with ETEC were the source of the outbreak. Chives were concluded to be the most likely source of infection, as they were imported from an ETEC-endemic country, not undergone any heat treatment before consumption and were not consumed immediately after being added to the cod, allowing growth of bacteria. The NFSA contacted the importer and distributors of the suspected chives and recommended holding back chives from the same batch and informing their clients about the suspicious contamination. In addition, NFSA planned to test newly imported chives from Kenya, but the importer stated they will not import any chives from Kenya before the summer.

Strong collaboration between different organisations (NIPH, NFSA and Veterinary institute) and different departments within each organisation enabled timely investigation and response to the outbreak. The outbreak was reported on Monday 14 March and on Friday 18 March preliminary results suggested cod with chives as possible source. On the same day chives were collected from the hotel and sent for microbiological analyses, simultaneously the importer was informed and recommended to keep back any chives belonging to the same batch. This shows the importance of good and timely communication and collaboration between all actors involved.

All ETEC, except for two patients, had different genetic profiles. A possible explanation could be that the chives were contaminated with multiple strains. In one patient (number 4 in table 4) multiple isolates were tested and the two ETEC strains identified showed different MLVA profiles. This suggests that this patient was infected with at least two different strains and support contamination of the chives with multiple strains of E coli.

This outbreak demonstrated the importance of complete knowledge of food preparation and serving. Fresh herbs and garnish are often associated with food borne outbreaks and therefore knowledge of all ingredients in the various dishes served is essential. Both in this and a previous ETEC outbreak in Norway (1), both dishes were cooked before the chives were added and the temperature of warm dishes could provide a good growth environment for ETEC introduced with the raw chives.

The NFSA recommends that commercial kitchens should heat-treat fresh herbs imported from outside Europe prior to serving (2). If heat treatment is not appropriate, the risk of growth of some infectious agents could be reduced by adding products right before consumption or serve them next

to the dish (2). This outbreak emphasizes the importance in following these guidelines to minimise risk of infections due to contaminated fresh herbs. In addition, it reinforces that imported fresh produce, including herbs, sprouts, lettuce and other vegetables should not be disregarded as possible vehicles for foodborne outbreaks in Norway, as well as other European countries.

References

1. MacDonald E, Møller KE, Wester AL, Dahle UR, Hermansen NO, Jenum PA, Thoresen L, Vold L. An outbreak of enterotoxigenic *Escherichia coli* (ETEC) infection in Norway, 2012: a reminder to consider uncommon pathogens in outbreaks involving imported products. *Epidemiol Infect.* 2015 Feb;143(3):486-93. doi: 10.1017/S0950268814001058. Epub 2014 May 9.
2. Mattilsynet website "Trygge frukt og grønnsaker er virksomhetens ansvar"
http://www.mattilsynet.no/mat_og_vann/produksjon_av_mat/frukt_bar_gronnsaker_og_korn/trygge_frukt_og_gronnsaker_er_virksomhetens_ansvar.7809