Impact factor 5.49

Eurosurveillance

Europe's journal on infectious disease epidemiology, prevention and control

Vol. 18 | Weekly issue 49 | 05 December 2013

Rapid communications

Surveillance and outbreak reports

ecdc EUROPEAN CENTRE FOR DISEASE PREVENTION AND CONTROL

www.eurosurveillance.org

Outbreak of *Salmonella enterica* Goldcoast infection associated with whelk consumption, England, June to October 2013

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Citation style for this article:
Inns T, Beasley G, Lane C, Hopps V, Peters T, Pathak K, Perez-Moreno R, Adak GK, Shankar AG, on behalf of the Outbreak Control Team. Outbreak of Salmonella enterica Goldcoast infection associated with whelk consumption, England, June to October 2013. Euro Surveill. 2013;18(49):pii=20654. Available online: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=20654

Article submitted on 22 November 2013 / published on 05 December 2013

An increase in the number of cases of *Salmonella enterica* serotype Goldcoast infection was observed in England during September 2013. A total of 38 cases were reported, with symptom onset dates between 21 June and 6 October 2013. Epidemiological, environmental, microbiological and food chain evidence all support the conclusion that this outbreak was associated with eating whelks processed by the same factory. Whelks are a novel vehicle of *Salmonella* infection and should be considered when investigating future outbreaks.

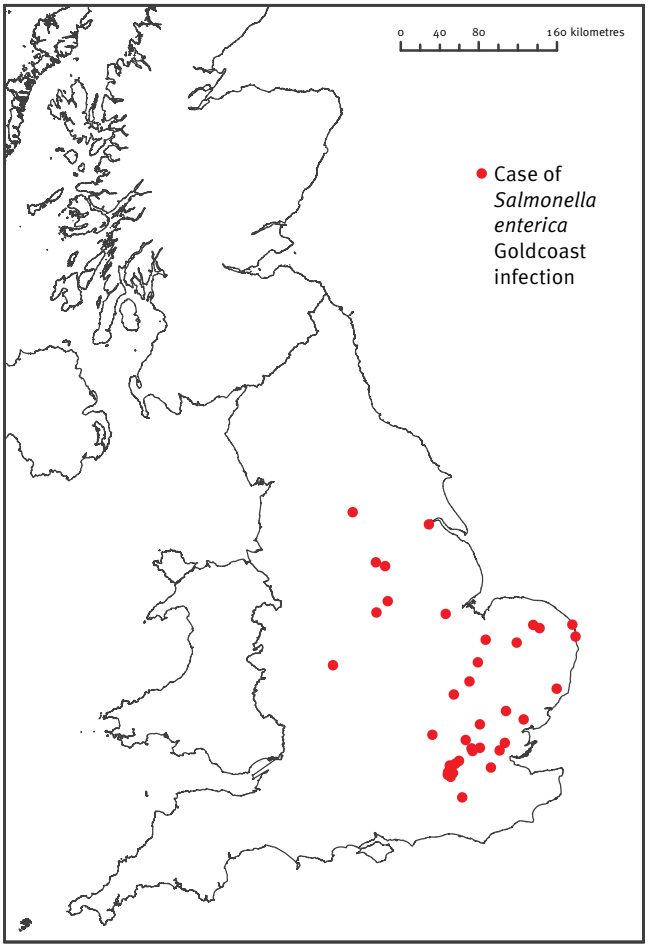
Identification of outbreak

During September 2013, 17 laboratory-confirmed cases of *Salmonella enterica* serotype Goldcoast infection with gastroenteritis were reported in England. This number was greater than the annually expected number: in 2012, eight cases were reported in England, in 2011, five were reported and in 2010, 13 were reported. Public Health England initiated an investigation on 12 September 2013 in order to identify the source of the outbreak and enable suitable control measures to be put in place to prevent further cases. The last case, with symptom onset in October, was reported on 12 November 2013, bringing the number of reported cases to 38. The cases' ages ranged between six months and 83 years (median: 64 years); two were aged under 16 years. Of the 38 cases, 25 were male. All cases were resident in England, predominantly in the east of the country (Figure 1).

As the incidence of this serotype is expected to be low, we looked at reports in the previous three months. Symptom onset dates ranged from 21 June 2013 (week 25) to 6 October 2013 (week 41) (Figure 2). Before this, the last case was reported in January 2013. Of the 38

Figure 1

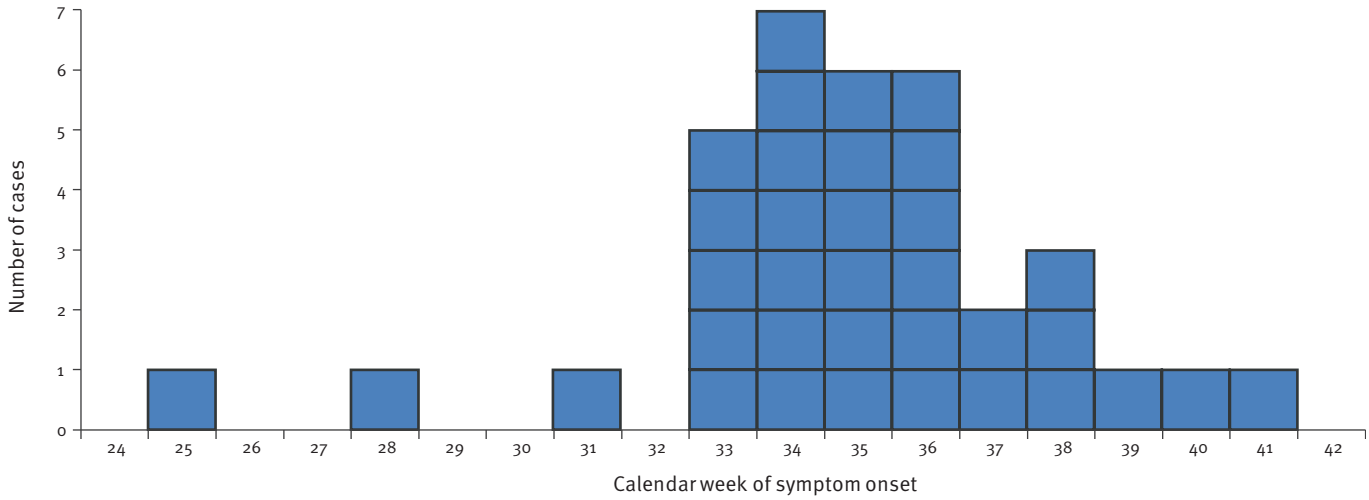
Place of residence of cases of *Salmonella enterica* Goldcoast infection, England, weeks 25–41[a] 2013 (n=38)



[a] Week 25 started on 17 June 2013.

Figure 2

Cases of *Salmonella enterica* Goldcoast infection, by calendar week[a] of symptom onset, England, weeks 24–42[a] 2013 (n=35)[b]



[a] Week 24 started on 10 June 2013.

[b] Onset dates were unobtainable for three cases.

cases, 10 were hospitalised, of whom four were admitted to intensive care. The median duration of illness was 13 days (range: 4–21); seven cases were symptomatic at the time they were first asked about their symptoms. Of the 35 cases for whom data on symptoms were available, all had diarrhoea; other symptoms included nausea (n=22), abdominal pain (n=22), fever (n=17), vomiting (n=15), headache (n=15) and blood in stools (n=3).

Epidemiological investigation

Outbreak cases were defined as persons resident in England diagnosed with *Salmonella* Goldcoast infection by the *Salmonella* Reference Service (Public Health England, London) after 1 June 2013. All laboratories in England and Wales send all *Salmonella* samples to this reference service.

To generate a hypothesis as to the source of this outbreak, we undertook detailed telephone interviews of the first 10 cases, with a trawling questionnaire. The questionnaire included demographics, clinical details, information on travel and contact with symptomatic persons, events attended and a detailed food history (including a general seafood question) for the week before symptom onset, including venues eaten at, and types and origin of foods eaten at home. Of the three cases with the earliest symptom onset dates, two reported having eaten whelks. Whelk consumption was reported by five of the 10 cases interviewed; this was much higher than the expected level of whelk consumption (for gastroenteritis questionnaires routinely completed in the east of England, eating whelks is reported by less than 1% of cases). Therefore, our primary hypothesis was that illness was associated with eating whelks (*Buccinum undatum*).

We then undertook an unmatched case–control study to test the hypothesis that whelk consumption was associated with *Salmonella* Goldcoast infection in England in 2013. Cases were excluded from the study if they were aged under 16 years (for logistic reasons), had travelled outside the United Kingdom (UK) in the five days before onset of symptoms, had had close contact with other individuals with gastroenteritis in the five days before onset, were asymptomatic or had already been interviewed with the trawling questionnaire. Controls were recruited through a systematic digit dialling process, using the home telephone number of cases to generate telephone numbers of people who were then contacted. We initially aimed to recruit two controls per case, but subsequently reduced this due to the strength of association observed. Information was collected from participants using a pre-tested questionnaire that was administered over the telephone by trained investigators. The questionnaire included questions on the following: demographics, clinical details, travel history, contact with persons with diarrhoea, events attended and foods eaten, including location of purchase, in the week before symptom onset. Foods in addition to whelks that were eaten by at least eight cases in the trawling questionnaire were included in the case–control study.

Data from the questionnaire were entered into a database using EpiData 3.1. Data were checked and analysed using Stata v12.1. The association between illness and each variable was estimated using odds ratios (ORs) and 95% confidence intervals (CIs). Data were subjected to univariable analysis and stratification to test for effect modification and confounding together with multivariable analysis using logistic regression.

Table

Univariable and multivariable associations between food exposure and *Salmonella enterica* Goldcoast infection, England, weeks 25–41[a] 2013

Food exposure	Case n=20[b]	Control n=27	Univariable analysis			Multivariable analysis		
	Number exposed	Number exposed	OR	95% CI	p value	OR	95% CI	p value
Whelks	16	1	104	9.54–4,517	<0.001	109	7.7–1,539	0.001
Cockles	5	0	NC	2.1–∞	0.010	–	–	–
Lettuce	10	22	0.2	0.05–0.99	0.030	–	–	–
Fish	7	18	0.3	0.07–1.06	0.420	–	–	–
Peppers	5	15	0.3	0.06–1.09	0.044	–	–	–

CI: confidence interval; NC: Not calculable; OR: odds ratio.

[a] Week 25 started on 17 June 2013.
[b] Two of the 22 cases eligible for inclusion in the case–control study chose not to participate.
[c] p value derived using Fisher's Exact test.

Statistically significant exposures at the alpha=0.05 level were added to the multivariable model. These were retained if they significantly improved the model, as assessed using a likelihood ratio test.

Food chain investigation

Foods identified in the trawling questionnaire and case–control study were investigated by environmental health officers and the Food Standards Agency. From each point of sale, distributers and suppliers were traced. Links between suppliers were mapped to produce a food chain diagram.

Food and environmental samples were taken using appropriate media. Samples were tested for presence of *Escherichia coli* and *Salmonella*. Presumptive *Salmonella* isolates were screened using a real-time polymerase chain reaction (PCR) assay for identification of the most common subspecies of *S. enterica* [1,2]. Serum agglutination tests, using Kauffmann–White classification [3], confirmed the presence of *S. enterica* serovar Goldcoast (6,8:r:l,w).

Analytical epidemiology

In total, 22 cases were eligible for the case–control study: of these, 20 cases were included (two cases declined to participate). A total of 27 controls were included. In a univariable analysis, cases were significantly more likely than controls to have consumed whelks, cockles, lettuce, fish and peppers (Table). No effect modification was detected. In the final multivariable model, when adjusted for sex, cases were significantly more likely to have consumed whelks (OR: 109; 95% CI: 7.7–1,539).

Food and environmental microbiology

Whelk consumption was reported by 24 of the 38 cases and one of the 27 controls; venues that these cases reported purchasing whelks from were investigated by environmental health officers. A summary of the food chain inferred from these investigations is shown in Figure 3. The supplier could be traced for whelks eaten by 20 cases: all were traced back to whelks processed by the same factory (Factory X). The whelks eaten by the control were not supplied by Factory X.

Factory X is a seafood factory in England; in 2012 it processed 639,049 kg of whelk meat. Whelks are processed by cooking in a pressure cooker. Their shells are then crushed and removed before the meat is cooled in a water bath. A small proportion is then sold fresh, with the majority of cooked whelks being flash frozen before sale. Over 90% of cooked whelks from Factory X are shipped to a single non-European Union country for further processing and consumption; the remainder are sold in the UK.

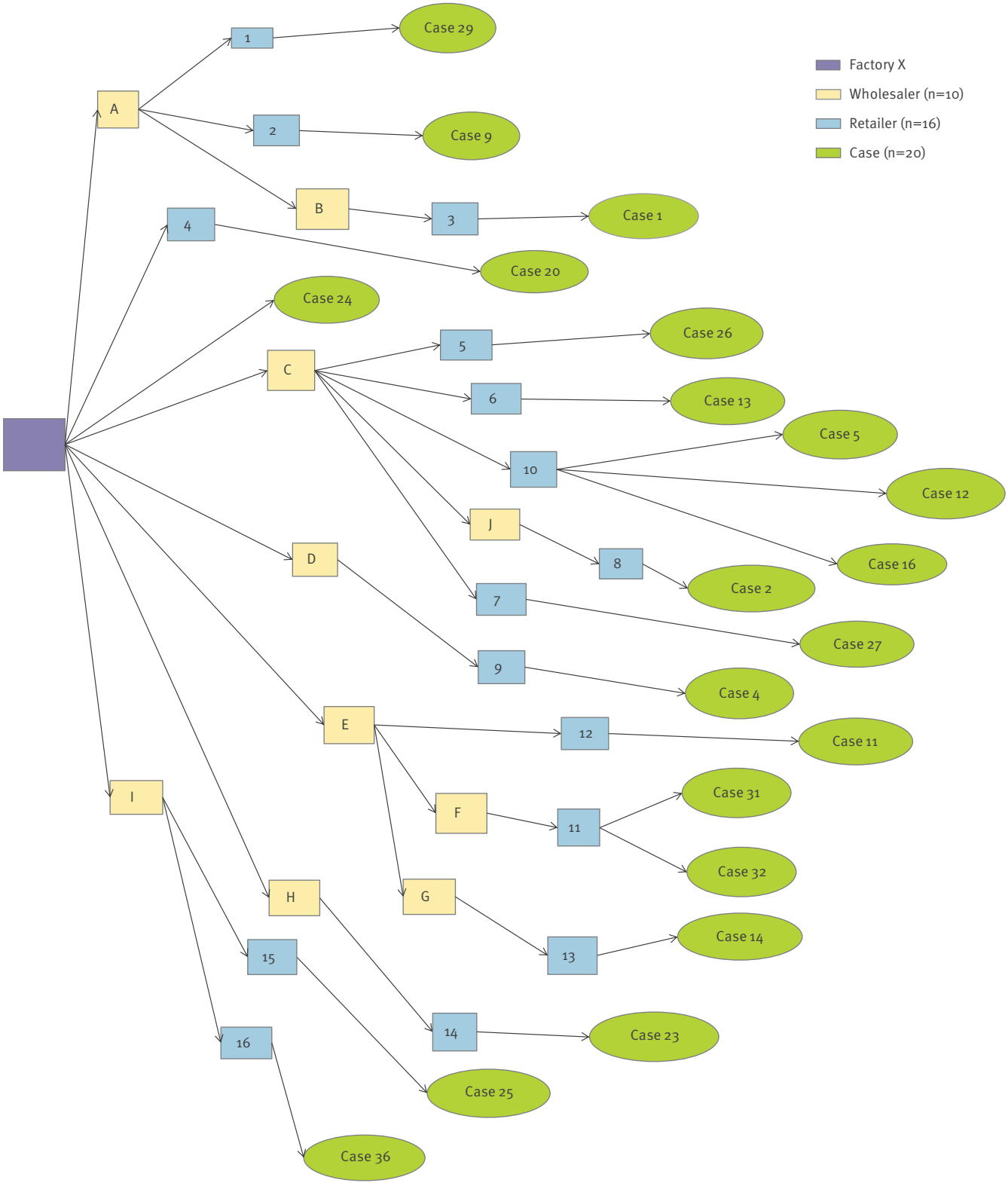
A total of 11 samples of whelks that had been processed at Factory X were taken at the point of sale from four outlets; two of the samples were positive for *Salmonella* Goldcoast. Seven processed and two raw whelk samples were taken from Factory X: none were positive for *Salmonella* Goldcoast. We tested 33 swabs or water samples from whelk-processing machinery at Factory X: six tested positive for *Salmonella* Goldcoast.

Control measures

Following initial descriptive epidemiology and food tracing investigations, Factory X was visited by environmental health officers on 20 September 2013. Cooking temperatures could not be verified and the factory agreed not to produce ready-to-eat whelks until further notice. During a subsequent inspection on 23 September, inadequate product temperatures were recorded immediately following cooking and therefore the whelks processed on that day and stored frozen on the premises were kept on the site. Due to the problems identified in the processing of whelks, the factory

Figure 3

Results of food chain investigations, *Salmonella enterica* Goldcoast infection, England, weeks 25–41[a] 2013 (n=20)



[a] Week 25 started on 17 June 2013.

instigated a product recall on 23 September 2013. The effectiveness of this recall was monitored by the Food Standards Agency and associated local authorities. The Foods Standards Agency informed the competent authorities in the country that this product is exported to: to date, no cases have been reported.

Discussion

We present epidemiological, environmental, microbiological and food chain evidence, which all support the conclusion that this outbreak of *Salmonella* Goldcoast infection was associated with consumption of whelks processed by Factory X. *Salmonella* Goldcoast outbreaks have previously been associated with pork products (salami [4], pork cheese (cooked pig organs stuffed in a pig stomach) [5], French paté [6] and raw fermented sausage [7]), watercress [8] and hard cheese [9]. Whelks have previously been associated with toxin-based food poisoning [10] but to our knowledge, this outbreak is the first known report of bacterial food poisoning associated with whelk consumption.

Regarding the mechanism of *Salmonella* Goldcoast contamination, microbiological evidence suggests that production equipment in contact with cooked whelks was contaminated with *Salmonella* Goldcoast for a number of weeks, despite the use of a sanitiser and the cooked whelks passing through a highly saline bath. *Salmonella* Senftenberg has previously been observed to survive in high salinity environments [11]: it may be that *Salmonella* Goldcoast shares this characteristic.

There was limited evidence that could indicate the original source of the *Salmonella* Goldcoast contamination. One of the environmental samples that tested positive for *Salmonella* Goldcoast was a swab of the conveyer belt used to transport raw whelks to the cooker, indicating that it was present on at least some whelks before entering the factory. Whelks are not filter feeders, and it is unclear whether they ingested the bacteria or the pathogens were in water that contaminated the shells. On the basis of the epidemiological evidence, we hypothesise that contaminated whelks may have been produced over at least a three-month period, but this contamination may have been intermittent, at a consistently low level. This would account for the relatively small number of cases seen.

We hope to be able to undertake whole genome sequencing of all outbreak cases and environmental isolates in future. Providing suitable background isolates are sequenced, this should allow a higher level of discrimination within the cases to ascertain which may be an artefact of sporadic incidence. Cases who did not report whelk consumption may have been sporadic or secondary cases. Regarding the case who was six months-old, one possible explanation is that the infant may have been infected by a family member who had eaten contaminated whelks, but who was asymptomatic.

Of the 38 cases, 10 were hospitalised, of whom four were admitted to intensive care. This level of severity has not previously been reported for *Salmonella* Goldcoast. *Salmonella* incidence is usually highest among the youngest (aged 0 to 4 years) [12]. As these cases had a median age of 65 years, they may have been more likely to have co-morbidities that increase the risk of hospitalisation.

One limitation of this study was that memory recall may have been different in case and control groups. To minimise this, cases and controls were interviewed in as timely a fashion as possible. Another limitation was that by using sequential digit dialling, a preponderance of females were recruited (20/27), whereas 25/38 of the cases were male. Explicit or frequency matching of controls was not possible due to the resources available for the investigation. To address this, sex was adjusted for in the analysis. It should also be noted that due to the small number of cases and controls, the CIs around the estimates of effect were wide.

We consider that the measures put in place to control this outbreak were effective in preventing further cases in England. Whelks are a novel vehicle of *Salmonella* infection and should be considered in the investigation of future outbreaks. It is known that processed whelks are sold internationally, and so if contaminated, there is the potential for cases to occur in countries outside the UK.

Acknowledgements

This article has been written on behalf of the Outbreak Control Team. With thanks to Anglia Health Protection Team administration and information staff for their work in the carrying out interviews for the case–control study, local Public Health England centres and Local Authorities for their assistance in the investigation of cases and Isabel Oliver, Richard Puleston and André Charlett for comments on the manuscript.

Conflict of interest

None declared.

Authors' contributions

All authors contributed to the writing of this manuscript and approved the final version.

TI designed the case–control study, analysed the data, drafted the manuscript and coordinated the outbreak response. GB organised data collection, food chain and environmental investigations. CL interpreted epidemiological and microbiological results. VH contributed to the environmental investigation. TP and KP were responsible for the microbiological results. RM coordinated the food chain investigation. GKA provided advice for the case–control study and the manuscript. AGS led and chaired the Outbreak Control Team, coordinated the outbreak response and advised on the manuscript.

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Tularaemia in a brown hare (*Lepus europaeus*) in 2013: first case in the Netherlands in 60 years

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Citation style for this article:
Rijks JM, Kik M, Koene MG, Engelsma MY, van Tulden P, Montizaan MG, Oomen T, Spierenburg MA, IJzer J, van der Giessen JW, Gröne A, Roest HJ. Tularaemia in a brown hare (Lepus europaeus) in 2013: first case in the Netherlands in 60 years. Euro Surveill. 2013;18(49):pii=20655. Available online: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=20655

Article submitted on 23 November 2013 / published on 05 December 2013

Tularaemia has not been reported in Dutch wildlife since 1953. To enhance detection, as of July 2011, brown hares (*Lepus europaeus*) submitted for post-mortem examination in the context of non-targeted wildlife disease surveillance, were routinely tested for tularaemia by polymerase chain reaction (PCR). *Francisella tularensis* subspecies *holarctica* infection was confirmed in a hare submitted in May 2013. The case occurred in Limburg, near the site of the 1953 case. Further surveillance should clarify the significance of this finding.

We report a brown hare (*Lepus europaeus*) infected with *Francisella tularensis*, the bacterium causing tularaemia, in the Netherlands in May 2013. This is the first case of tularaemia in Dutch wildlife since 1953. The finding results from the intensified surveillance of the disease in brown hares, which started in July 2011 after infected hares were detected in neighbouring countries.

Tularaemia, caused by the bacterium *F. tularensis*, is a zoonotic disease that was reported in more than 800 humans in the European Union in 2010 [1]. The bacterium has a very wide host range that includes mammals, birds, amphibians, fish, and invertebrates [2]. It can remain infectious in water and mud for months [3]. It can be transmitted by inhalation of infective aerosols, contact with or ingestion of infected hosts or water, and arthropod bites [2].

Four subspecies can be distinguished: *tularensis*, *holarctica*, *mediasiatica* and *novicida*. The first two subspecies are important causes of tularaemia in humans and animals [2]. The bacterium has been detected in wildlife in various European countries, such as Denmark, Finland, France, Germany, Spain, Sweden and Switzerland in 2012; Belgium, Italy and Norway in 2011; and Austria in 2009 (Figure 1) [4].

Lagomorphs and rodents are most susceptible to infection and disease by the bacterium [5]. In a number of European countries, brown hares are considered to be an important host of *F. tularensis* and transmission to humans is known to result from direct contact with hares [6,7].

In the Netherlands, the agent was last reported in 1953 when seven members of an eight-person family became ill after consuming a brown hare [8,9]. In contrast, in Lower Saxony, a German federal state that shares a common border with the Netherlands, *F. tularensis* has been detected in 2.9% of hares found dead as recently as in the period between 2006 and 2009 [10]. In addition, in the autumn of 2011 tularaemia was found in hares in Düren, a municipality in North Rhine-Westphalia, Germany, about 50 km from the eastern Dutch border [11], as well as in Anthisnes, a municipality in the Province of Liege, Belgium, approximately at the same distance from the southern Dutch border [12].

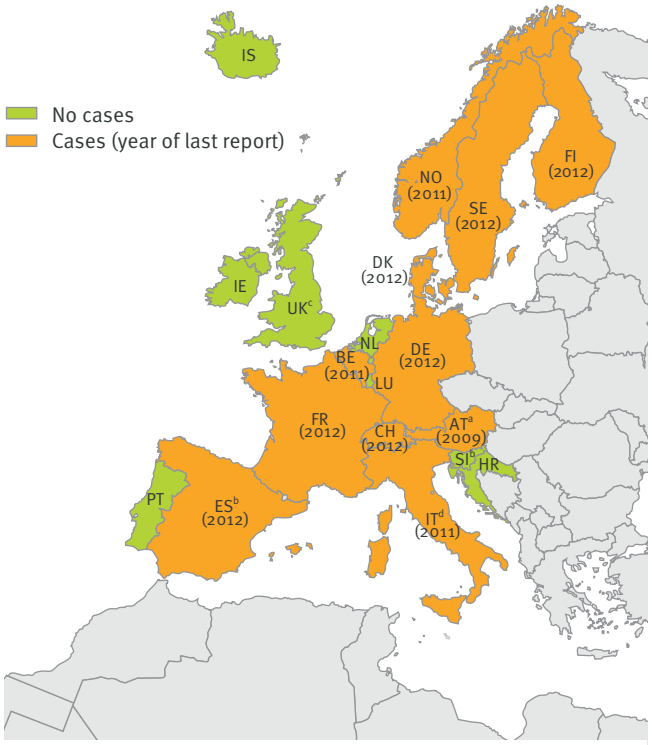
These recent reports suggested that *F. tularensis* may also be present in the Netherlands without being detected. Therefore the Dutch Wildlife Health Centre (DWHC) and the Central Veterinary Institute (CVI) in collaboration with the National Institute for Public Health and the Environment (RIVM) decided to intensify surveillance for tularaemia in brown hares in the Netherlands.

Finding of a brown hare testing positive for *Francisella tularensis*

Brown hares that are found either dead or terminally ill and then euthanised by hunters or game wardens can be submitted to the DWHC for post-mortem examination in the context of non-targeted surveillance. Since July 2011, these hares have been routinely tested for the presence of *F. tularensis* DNA by polymerase chain reaction (PCR) at CVI. DNA was extracted from lung and/or spleen using a DNA tissue kit (DNeasy Blood and Tissue Kit; Qiagen, Hilden,

Figure 1

Cases of tularaemia in animals in some European Union Member States, Croatia, Iceland, Norway and Switzerland, 2006–2012 (n=19 countries)



AT: Austria; BE: Belgium; CH: Switzerland; DE: Germany; DK: Denmark; ES: Spain; FI: Finland; FR: France; HR: Croatia; IE: Ireland; IS: Iceland; IT: Italy; LU: Luxembourg; NL: Netherlands; NO: Norway; PT: Portugal; SE: Sweden; SI: Slovenia; UK: United Kingdom.

[a] No information on the occurrence of tularaemia in animals was provided to the World Animal Health Information Database (WAHID) for AT before and after the year 2009.

[b] No information on the occurrence of tularaemia in animals was provided to WAHID for ES and SI in 2006.

[c] No information on the occurrence of tularaemia in animals was provided to WAHID for the UK for 2006 to 2007.

[d] No information on the occurrence of tularaemia in animals was provided to WAHID for IT for 2006 to 2008.

Source: The map was compiled based on information from the WAHID accessed on 20 August 2013.

Germany). The extracted DNA was tested by TaqMan real-time PCR using the FTT0376 primers and probe published by Mitchell et al. [13] (forward primer 5'-CCATATCACTGGCTTTGCTAGACTAGT-3', reverse primer 5'-TGTTGGCAAAAGCTAAAGAGTCTAAA-3', probe 5'-FAM-AAATTATAAAACCAAACCCAGACCTTCAAACCACA-BHQ1-3'). This assay is specific for the pathogenic subspecies of *F. tularensis* (subspecies *tularensis*, *holarctica* and *mediasiatica*). The positive samples were also sent to the Swedish Veterinary Institute (SVA), Uppsala, Sweden for confirmation.

By May 2013 a total of 49 animals from nine of the 12 Dutch provinces had been examined (Figure 2). Of the 49 specimens, 26 had one or more macroscopic or microscopic lesions consistent with tularaemia in this species [14]. The first 48 hares tested negative for the presence of *F. tularensis* DNA by PCR. The 49th hare examined in May 2013 was an adult male hare from the province of Limburg. Prior to death, the animal had been seen with an unsteady gait, had been reluctant to move and was easy to catch.

The slightly autolytic carcass of this animal had an enlarged spleen at necropsy and histopathology revealed multiple foci of hepatocellular necrosis, consistent with *F. tularensis* infection [14]. Real-time PCR analyses of spleen and lung samples of this specimen were positive for *F. tularensis*. Culture of the samples from this animal on chocolate agar medium with cysteine and sodium sulphite provided negative results. Infection by *F. tularensis* was confirmed at the SVA, both by PCR (spleen) and immunohistochemistry (lung). The subspecies of *F. tularensis* was subsequently typed by CVI as *holarctica* based on the concatenated partial sequences of five metabolic housekeeping genes as described by Nübel et al. [15].

Discussion and conclusion

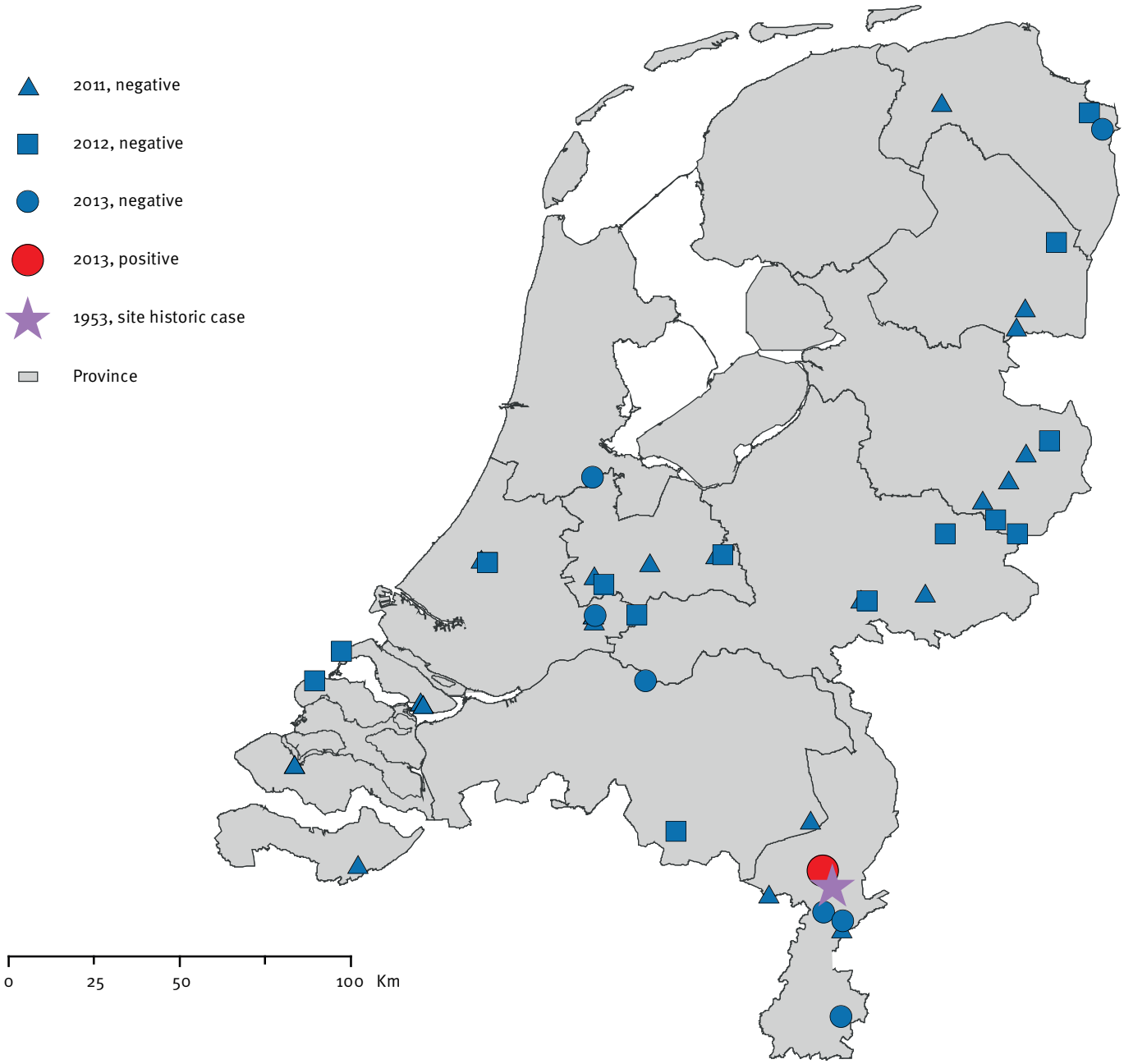
F. tularensis ranks among the top twenty emerging zoonotic pathogens considered to be relevant for the Netherlands [16]. The emergence or re-emergence of the disease in other countries has been associated with factors such as climate change, human-mediated movement of infected animals, as well as with conditions of war with subsequent increase in rodent populations. In some cases, detection due to enhanced surveillance revealed the presence of the disease [2,17]. Enhanced surveillance also likely contributed to the apparent re-emergence of tularaemia in Dutch wildlife after 60 years, as reported here.

The subspecies *F. tularensis holarctica* detected in this study is consistent with the subspecies detected in wildlife in the neighbouring countries [10]. The infected hare was found only 6 km away from the home of the family in the 1953 report [8,9] (Figure 2). It is unclear as to how widespread the occurrence of the bacterium is in wildlife in the Netherlands and therefore whether the proximity of both events indicates a hot spot or a coincidence. Heightened surveillance is needed in order to answer this question.

Given the proximity of these cases to the border, emergence due to import of the disease from neighbouring countries should also be considered. Indeed, in 2012 four cases of tularaemia in hares were identified in the area of Heinsberg, Hückelhoven and Erkelenz in Germany, 10 km from the Dutch border and 30 km from the case reported here [18]. It is unlikely that hares are deliberately introduced from abroad into the Netherlands, since release of hares is illegal. It is also unlikely that the specific hare found infected in this

Figure 2

Location and year of sampling of hares with polymerase chain reaction test results for *Francisella tularensis* (n=49) relative to where an infected hare was found in 1953, Netherlands, 2011–2013



In the legend, the terms 'negative' and 'positive' are used to describe the sampled hares' polymerase chain reaction test results for *Francisella tularensis*.

study came from abroad on its own, as hares do not usually cover such distances and in addition, a large river separates the two locations [19,20]. However, the infection may have moved more gradually into the Dutch area through other hosts or vectors or both without having been detected.

Only two human tularaemia cases likely to be autochthonous have been recorded in the Netherlands since 1953, though human infection was notifiable from 1976 to 1999 [9,21,22]. One case occurred in 2011 and one in 2013, and neither had a history of contact with dead hares or other animals [21,22]. However, it is presumed that the 2013 case may have contracted the disease in Limburg through insect bites [22]. Both, these human cases and the hare case, highlight the importance of raising the awareness of physicians and veterinarians with regards to the disease.

These findings support the continuation of non-targeted disease surveillance in hares and other wildlife species, and heightened targeted surveillance for tularaemia, with focus on the affected regions. Hunters, game wardens, and other groups that are likely to come into close contact with wildlife will be informed and included in these activities.

Acknowledgements

The authors thank Y Bisselink, R Ruuls, N Buijs, and M Buitelaar for technical and administrative assistance, SVA for confirmation testing by PCR and immunohistochemistry, and R Thomas for her suggestions for textual corrections. Funding statement: Non-targeted surveillance of wildlife in the Netherlands was funded by the Faculty of Veterinary Medicine of Utrecht University, the Dutch Ministry of Economic Affairs and the Dutch Ministry of Health, Welfare and Sport. Laboratory testing at CVI was primarily financed by the Ministry of Economic Affairs through the project 'meldingsplichtige ziekten', grant number WOT-01-002-05.02. Confirmation at SVA was funded by the Wildtech project (EU 7th Framework Program for Research and Technological Development, grant agreement no. 222633).

Conflict of interest

None declared.

Authors' contributions

JMR, MGM, JWvdG, TO, MAS, AG, HJR were involved in the surveillance set-up and its general implementation. MK and JIJ performed the post-mortem examinations; PvT, MYE and MGK the diagnostic tests. JMR, AG, MYE and HJR drafted the manuscript, and MK, MGK, PvT, MGM, JIJ and JWvdG critically revised it. JMR provided figures 1 and 2. All authors approved the final version of the manuscript.

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Case of vaccine-associated measles five weeks post-immunisation, British Columbia, Canada, October 2013

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Citation style for this article:
Murti M, Krajden M, Petric M, Hiebert J, Hemming F, Hefford B, Bigham M, Van Buynder P. Case of vaccine-associated measles five weeks post-immunisation, British Columbia, Canada, October 2013. Euro Surveill. 2013;18(49):pii=20649. Available online: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=20649

Article submitted on 15 November 2013 / published on 05 December 2013

We describe a case of vaccine-associated measles in a two-year-old patient from British Columbia, Canada, in October 2013, who received her first dose of measles-containing vaccine 37 days prior to onset of prodromal symptoms. Identification of this delayed vaccine-associated case occurred in the context of an outbreak investigation of a measles cluster.

In this report we describe a case of measles-mumps-rubella (MMR) vaccine-associated measles illness that was positive by both PCR and IgM, five weeks after administration of the MMR vaccine. Based on our literature review, we believe this is the first such case report which has implications for both public health follow-up of measles cases and vaccine safety surveillance.

Between 29 August and 2 September 2013, three unlinked persons from across the Fraser Valley, British Columbia, Canada, presented with rash illness consistent with clinical measles [1]. Based on the outbreak investigation by the local health authority, none of the three cases had an identified exposure to a measles case or travel history outside of Canada during the incubation period, and a source case was never identified. All three cases had the same measles genotype B3 sequence type (MVs/British Columbia.CAN/34.13, MeaNS id 39928, GenBank accession numbers KF704002 and KF704001). Measles genotype B3 is endemic in the World Health Organization's African and Eastern Mediterranean regions [2]. Two additional cases of measles due to secondary transmission from one of the above cases were identified in British Columbia in the third week of September.

Case report

In early October 2013, a two-year-old child living in the Fraser Valley presented to the family physician with fever, rash, conjunctivitis and coryza. Symptoms had begun two days before, with a runny nose, followed by fever on the day hereafter. A macular rash appeared on the day of visiting the physician, starting on the face and progressing to the rest of the body; fever measured by the parents was at 39 °C.

Clinical examination of the child by the family physician found a fever of 39.5 °C, marked bilateral conjunctivitis, and macular rash over the body. Three days later, fever had dissipated, rash was fading and symptoms resolved without complications.

Public health alerts had been issued to community physicians regarding the recent cluster of measles in September, which may have raised suspicion for measles in this case. Additionally, the child's family was aware of measles cases in the community from a relative who attended the same church as one of the original cases, but no direct link was identified and they had no travel history outside of Canada. Contact investigation revealed no ill household members or preschool contacts. The child's past medical history indicated anaphylaxis to peanuts and eggs. Primary series of immunisations were not up-to-date, as she had just received her first dose of MMR vaccine 37 days prior to the onset of illness. At the same visit, the child had received meningococcal C and pneumococcal conjugate vaccines.

Laboratory investigations

Laboratory testing for measles was performed on specimens collected on the day of rash onset. Measles RNA was detected in the nasopharyngeal swab by the RT-PCR assay [3]. Acute and convalescent measles specific IgM and IgG antibodies were detected in the blood by ELISA (Enzygnost Anti-Measles Virus IgM and IgG (Dade Behring, Marburg, Germany): IgM detectable (0.213), IgG 1294 mIU/mL, and IgM detectable (0.246), IgG 2,413 mIU/mL, respectively. Virus genotype was determined by the National Microbiology Laboratory in Winnipeg, Canada as vaccine strain, genotype A, MVs/British Columbia/39.13 [A] (VAC) [4]. Other virology testing found no detectable Parvovirus B19 specific IgG or IgM antibody, and detectable human herpesvirus

(HHV)-6 specific IgG antibody but no detectable HHV-6 DNA.

Public health measures

While genotyping results were pending, case management proceeded as for a wild-type measles infection. Public health follow-up lead to the identification of 87 contacts. As per guidelines, post-exposure prophylaxis was provided within six days of exposure to 45 susceptible contacts (41 contacts with a history of one dose of MMR vaccine received an additional MMR dose, and four contacts with no history of MMR vaccine or with contraindications to MMR vaccination, received immunoglobulin) [1]. All contacts received education on signs and symptoms of measles, and those who received immunoglobulin were recommended to subsequently receive MMR vaccine, if this was not contraindicated.

Discussion

The incubation period of measles is typically eight to 12 days from exposure to rash onset, with a range from seven to 21 days. Public health interventions are based on this established incubation period for determining the epidemiological links between cases and for estimating periods of exclusion for contacts in high risk settings [5,6]. Based on our review of the literature, this report documents the first case of MMR vaccine-associated measles, 37 days post-immunisation, well beyond 21 days and the routine 30 days post-MMR immunisation period used by the Canadian adverse event following immunization (AEFI) surveillance system.

Measles-containing vaccines are used globally, have been part of the British Columbia immunisation schedule since 1969, and have an impressive record of safety validated by careful, ongoing AEFI surveillance. Rash and/or mild clinical illness following MMR vaccine are not uncommon [7]. Clinically significant vaccine-associated illness is rare, but when it occurs it is indistinguishable from wild-type measles, except by genotyping [8]. Detection of vaccine virus has been documented up to 14 days post-immunisation by RT-PCR, and up to 16 days by immunofluorescence microscopy of urine sediment [9-12]. Complications from vaccine-associated measles have been documented in both immune-competent and compromised individuals [13,14]. Of note, only one case report of transmission from vaccine-associated measles has been identified [15,16].

Possible explanations for this prolonged shedding of measles vaccine virus include interference with the immune response by host or vaccine factors. Immunoglobulin administration early in the incubation period has been reported to extend the time to onset of symptoms, but in this child there was no such history and no known immunosuppressive illness [5]. The two-fold rise between acute and convalescent measles-specific IgG suggests the vaccine-mediated immune response had been underway prior to the onset of symptoms. Investigations clarified that there were no shipping, handling or cold-chain deviations for the specific vaccine used, and that it was administered by a public health nurse trained in immunisations. The potential immunological impact of the older age of the child at the time of receiving the first dose of MMR vaccine, 33 months versus the typical 12-15 months of age, and the co-administration of meningococcal C and pneumococcal conjugate vaccines are areas for future investigation.

It is possible that the case's symptoms were not measles-vaccine-related but an inter-current illness confounding the presentation. However, symptoms of marked conjunctivitis, continued fever with rash, and progression of macular rash from face to the whole body, are all more suggestive of measles versus other exanthems caused by viral diseases. Parvovirus and HHV-6 results were negative, and the absence of intake of medications excludes a drug reaction. Rubella serology was not done as it was expected to be positive given the recent MMR vaccine administration. Therefore, the combination of classic measles symptoms, detection of measles vaccine virus and reactive measles IgM, and lack of evidence of an alternative illness explanation, were highly suggestive of measles vaccine-associated illness.

Heightened surveillance and awareness of measles because of the ongoing outbreak likely contributed to the identification of this case. Although this is the first such reported case, it likely represents the existence of additional, but unidentified, exceptions to the typical timeframe for measles vaccine virus shedding and illness. Such cases have important public health implications for the investigation of measles clusters because while there is uncertainty about case classification (wild-type vs vaccine-type), case and contact management should proceed as if for wild-type to prevent secondary transmission. In this case, uncertainty from the presence of a measles outbreak, symptom onset on day 37 after MMR vaccine administration, and a two-week period between the RT-PCR findings and genotype determination, resulted in the initially reasonable presumption that this was a wild-type measles case and subsequent resource-intense follow-up of contacts. Awareness of the frequency of such exceptions to the typical measles timeframe and improving the timeliness of measles vaccine virus genotyping could help focus public health resources on cases of wild-type measles. Further investigation is needed on the upper limit of measles vaccine virus shedding based on increased sensitivity of the RT-PCR-based detection technologies and the immunological factors associated with vaccine-associated measles illness and virus shedding.

Acknowledgements

We would like to thank the patient and their family, as well as the FHA public health nurses, Stephanie Konrad, Monika Naus, and David Puddicombe for their assistance.

Conflict of interest

None declared.

Authors' contributions

BH, FH, MM and PVB contributed to the clinical and public health management of the case. MK, MP and JH provided laboratory testing. MM drafted the manuscript; all authors critically revised and approved the final version of the manuscript.

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A *Shigella sonnei* outbreak traced to imported basil – the importance of good typing tools and produce traceability systems, Norway, 2011

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Citation style for this article:
Guzman-Herrador BR, Nilsen E, Cudjoe KS, Jensvoll L, Kvamme JM, Lindegård Aanstad A, Lindstedt BA, Nygård K, Severinsen G, Werner-Johansen Ø, Wester AL, Wiklund M, Vold L. A Shigella sonnei outbreak traced to imported basil – the importance of good typing tools and produce traceability systems, Norway, 2011. Euro Surveill. 2013;18(49):pii=20650. Available online: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=20650

Article submitted on 30 November 2012 / published on 05 December 2013

On 9 October 2011, the University Hospital of North Norway alerted the Norwegian Institute of Public Health (NIPH) about an increase in *Shigella sonnei* infections in Tromsø. The isolates had an identical 'multilocus variable-number tandem repeat analysis' (MLVA) profile. Most cases had consumed food provided by delicatessen X. On 14 October, new *S. sonnei* cases with the same MLVA-profile were reported from Sarpsborg, south-eastern Norway. An outbreak investigation was started to identify the source and prevent further cases. All laboratory-confirmed cases from both clusters were attempted to be interviewed. In addition, a cohort study was performed among the attendees of a banquet in Tromsø where food from delicatessen X had been served and where some people had reported being ill. A trace-back investigation was initiated. In total, 46 cases were confirmed (Tromsø= 42; Sarpsborg= 4). Having eaten basil pesto sauce or fish soup at the banquet in Tromsø were independent risk factors for disease. Basil pesto was the only common food item that had been consumed by confirmed cases occurring in Tromsø and Sarpsborg. The basil had been imported and delivered to both municipalities by the same supplier. No basil from the specific batch was left on the Norwegian market when it was identified as the likely source. As a result of the multidisciplinary investigation, which helped to identify the source, the Norwegian Food Safety Authority, together with NIPH, planned to develop recommendations for food providers on how to handle fresh plant produce prior to consumption.

Introduction

Shigellosis is endemic throughout the world. Symptoms are usually mild but range from watery, self-limiting diarrhoea to life threatening dysentery [1]. Of four *Shigella* species, *S. sonnei* is the most frequently isolated in industrialised countries [2]. Symptoms of *S. sonnei* infection are usually milder than those caused by *S. dysenteriae* or *flexneri* [3]. The bacteria are transmitted by ingestion of contaminated food or water, or through person-to-person contact. The incubation period ranges from 12 hours to one week [4]. The infective dose is very low: ingestion of 100 to 200 microorganisms can lead to disease [3].

In the European Union (EU) shigellosis infections are relatively uncommon. With a rate of 1.64 cases per 100,000 population in 2010, they are far less frequent than *Campylobacter* and *Salmonella* infections, which have respective incidences of 56.95 and 21.31 per 100,000 population [5]. In Norway, between 120 and 190 cases of shigellosis have been reported annually in the last ten years, corresponding to an incidence between 2.5 to 4.0 per 100,000 population. Only 10 to 20% of the cases are domestically acquired [6]. Imported fresh vegetables have been identified as the vehicle of several outbreaks over the last years [7,8].

In order to control imported feed and food of non-animal origin, the European Commission Regulation (EC) No 669/2009 specifies a list of risk products subjected to increased level of official controls upon entry into

the European Economic Area, which includes Norway [9].

The alert

On 8 October 2011, clinicians at the University Hospital of North Norway in Tromsø, northern Norway (2011 population: 68,200 inhabitants) [10], attended six patients with bloody diarrhoea. On 9 October, the hospital's Department of Microbiology and Infection Control confirmed three patients with *S. sonnei* infection. None of them had a travel history outside or within Norway in the previous week. The microbiologist on call reported the cluster to the Municipal Medical Officer in Tromsø and to the Department of Infectious Diseases Epidemiology at the Norwegian Institute of Public Health (NIPH), and isolates were forwarded to the National Reference Laboratory for Enteropathogenic Bacteria (NRL). The NRL verified the isolates as being *S. sonnei* with an identical multilocus variable-number tandem repeat analysis (MLVA) profile that had not been identified in Norway before. Concurrently, the Local Food Safety Authority in Tromsø interviewed the patients on food consumption, who reported having eaten at delicatessen X in downtown Tromsø or having participated in social events with food provided by delicatessen X during the week before becoming sick. In addition, the owners of delicatessen X, who were also interviewed, had received complaints from customers who had fallen ill.

On 14 October, the Municipal Medical Officer in Sarpsborg (2011 population: 52,800) [10], 1,700 km south of Tromsø, notified a second cluster of shigellosis, whereby none of the patients had a travel history to Tromsø. *S. sonnei* isolates had the same MLVA profile as those from Tromsø.

Since more than one county was affected, the further coordination of the investigation was transferred to the national level. NIPH, in collaboration with the Norwegian Veterinary Institute, the Norwegian and Local Food Safety Authorities, the Municipal Medical Officers of the municipalities involved, and the Department of Microbiology and Infection Control at the University Hospital of North Norway, investigated the outbreak to identify the source, implement control measures and prevent further cases.

Methods

Epidemiological investigation

A case was defined as (i) a person in Norway with laboratory-confirmed *S. sonnei* infection after 1 October 2011 with the MLVA profile identified in the outbreak with absence of travel history abroad, or (ii) a person who had an isolate with one-locus difference from the MLVA outbreak profile and an epidemiological link to (i). The Local Food Safety Authority interviewed cases in Tromsø by telephone using a standard food-borne disease trawling questionnaire to generate hypotheses about common exposures among cases. Once the suspicion was focused towards Delicatessen X, their menu was used as basis for the interviews.

In order to gather more information on the second cluster, NIPH interviewed all cases in Sarpsborg by telephone using the same food-borne disease trawling questionnaire looking for common exposures among them and to those in Tromsø.

Cohort study in Tromsø

Delicatessen X provided a list of social events they catered for from 30 September to 8 October, including a banquet with 50 guests in Tromsø on 1 and 2 October. Since the organiser of the banquet had reported to the delicatessen that some of them had fallen ill, and this event included participation of a greater number of participants, the NIPH studied a cohort among the banquet attendees to identify risk factors for disease. For the cohort study we defined a 'probable case' as a person who developed diarrhoea (more than three loose stools in 24 hours) and fever (self-reported) up to seven days after the banquet.

On 14 October, the NIPH sent a link to a web-based questionnaire via e-mail to the attendees. It contained questions on demographic information, symptoms and food eaten. NIPH attempted to interview persons who had not replied within five days by phone. Attack rates and relative risks with 95% confidence intervals (CI) were calculated. Variables with $p<0.1$ in the univariate analysis were included in a multivariable logistic regression model, using. STATA 11.0 (Stata Corporation, College Station, TX, USA).

Microbiological investigation

The Department of Microbiology and Infection Control at University Hospital of North Norway identified the initial isolates as *S. sonnei* by fermentation tests, agglutination and Vitek 2 automated identification. They sent them to the NRL, where all *Shigella* spp. isolates identified in Norway are received for identification to species-level, O-serogrouping, antimicrobial resistance testing and MLVA-typing [11].

Table 1

Description of the laboratory-confirmed cases of *Shigella sonnei* infection in Tromsø (n=42) and Sarpsborg (n=4), Norway, October 2011

Variable	Tromsø	Sarpsborg
Sex		
Female	24	3
Male	18	1
Age (years)		
Median	41	46.5
Range	19–84	45–64

The Norwegian Veterinary Institute analysed, using a polymerase chain reaction (PCR)- based method elaborated by the Nordic Committee on Food Analysis, the food items served by Delicatessen X in Tromsø and those served in Sarpsborg. Specimens were analysed for *Shigella* spp., *Enterobacteriaceae* and thermotolerant coliforms [12-14].

Trace-back investigation

The Norwegian Food Safety Authority performed trace back of products and inspection of the premises where the food likely associated to this outbreak was distributed, prepared and served. The supplier of the relevant food items was contacted to document and provide an overview of the supply chain process.

Results

Epidemiological investigation

Forty-six cases with identical MLVA profile were reported: 42 cases linked to Tromsø and four to Sarpsborg (Table 1). None of them reported travel outside Norway during the week prior to the onset of symptoms.

Of the Tromsø cases, all were diagnosed in Tromsø, with the exception of one who was diagnosed in Oslo, but reported travel to Tromsø in the previous week (Figure 1). The cases in Tromsø had isolates collected and tested for gastrointestinal pathogenic bacteria between 5 and 21 October and those in Sarpsborg between 11 and 25 October (Figure 2).

Cluster in Tromsø

The median age of all 42 cases in Tromsø was 41 years (range: 19–84 years); twenty-four of the cases were female (Table 1). The first case of the outbreak sought medical attention and was tested on 5 October and the last one on 21 October (Figure 2). In total, four patients were hospitalised. All of them were admitted during the first days of the outbreak and had bloody diarrhoea; fever and abdominal pain, with a mean C-reactive protein of 234 mg/L (range: 120–364 mg/L; norm <10mg/L). The mean length stay in hospital was 2.8 days (range: 1–4 days). Three of the hospitalised patients received antibiotic treatment and all the admitted patients recovered well.

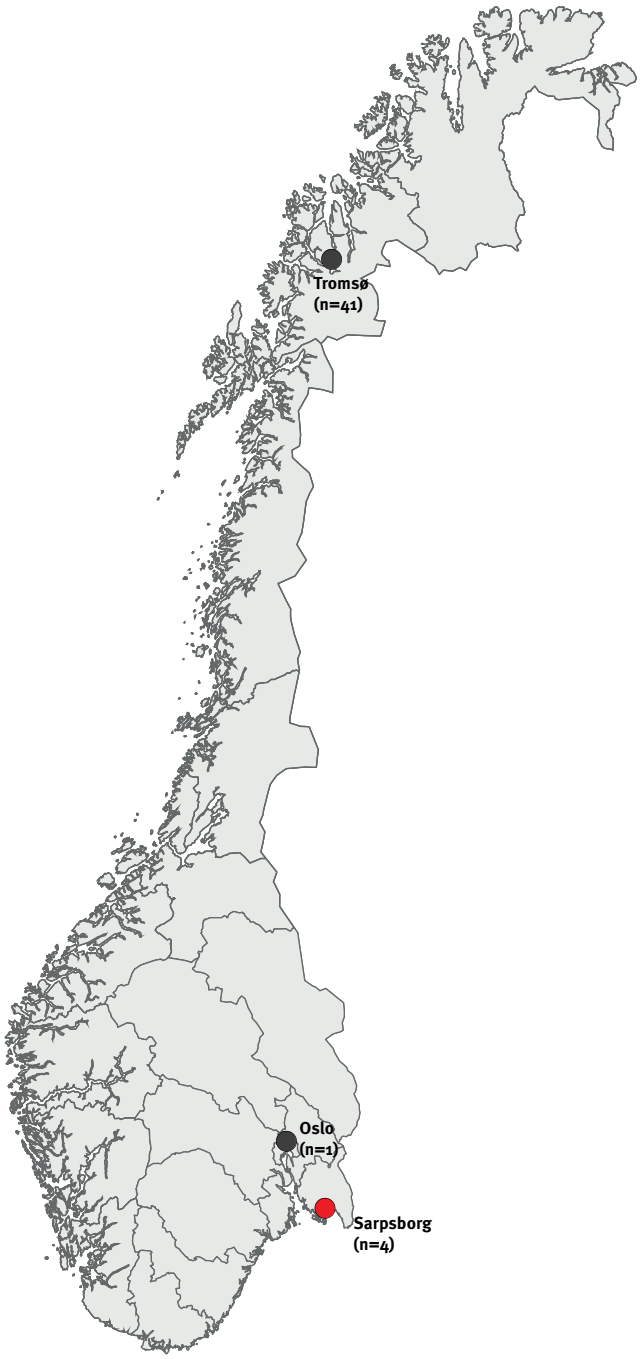
The Local Food Safety Authority interviewed 38 of the 42 cases: 37 had eaten food containing pesto sauce made with fresh basil from the Delicatessen X.

Cohort study in Tromsø

Forty-two of the fifty banquet attendees answered the web-based questionnaire. Eleven met the probable case definition (attack rate: 26%). All of them had diarrhoea. Frequent symptoms were also abdominal pain (8 persons) and fever and nausea (7 persons). The highest attack rate occurred among those aged 20 to 29 years (4/11; 36%) although there were persons from all ages affected. Both sexes were equally affected.

Figure 1

Geographical distribution of *Shigella sonnei* infection cases by municipality of diagnosis, Norway, October 2011



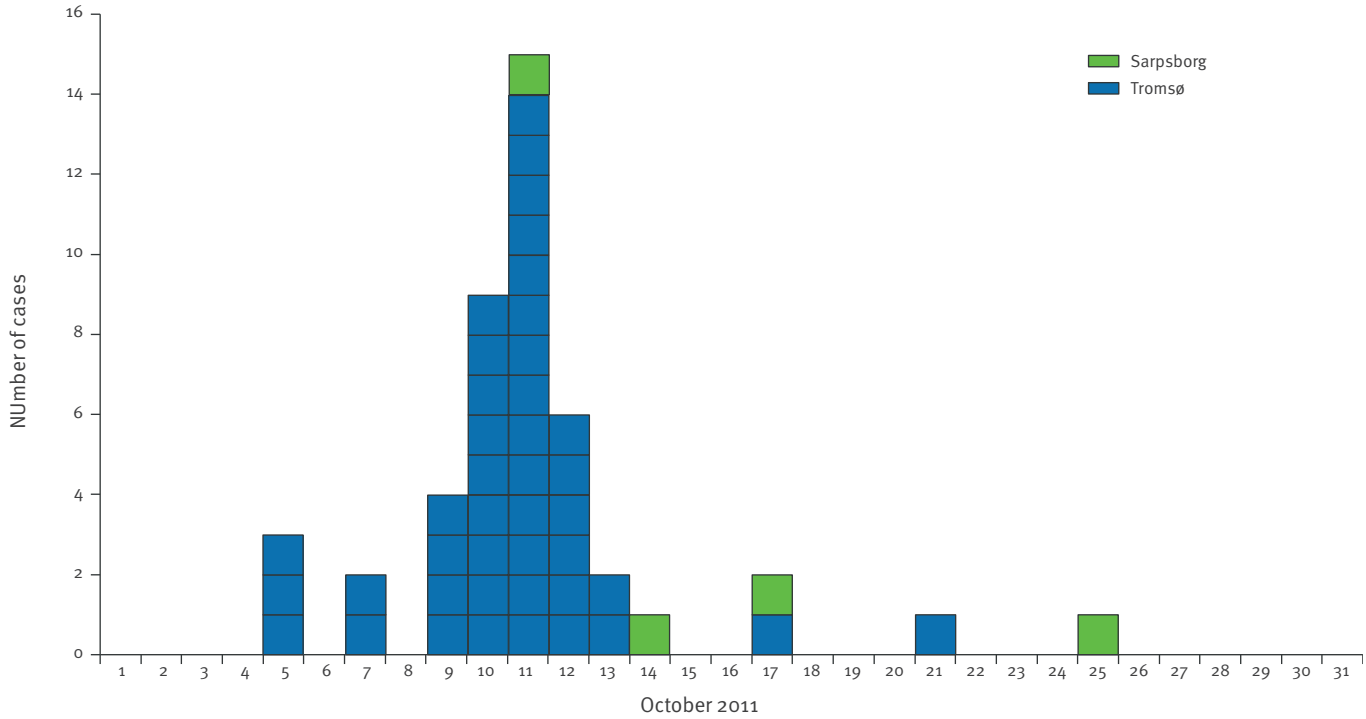
The case diagnosed with *Shigella sonnei* infection in Oslo, reported travel to Tromsø in the week before being diagnosed.

Six probable cases sought medical attention and three of them had a stool sample taken and were laboratory confirmed with the *S. sonnei* outbreak strain.

Ten banquet food items were significantly associated with disease in the univariate analysis (Table 2). Attendees exposed to basil pesto sauce had the

Figure 2

Cases of *Shigella sonnei* infection by date of specimen collection, Norway, October 2011 (n=46)



highest attack rate (78%). Among the three food items with the highest relative risks (RR), basil pesto sauce had the smallest confidence interval. The lower limit of the confidence interval was higher than for any other item (RR: 5.4; 95% CI: 2.1–14.4).

Only two food items, fish soup (odds ratio (OR): 8.2; 95% CI: 1.1–61.1) and basil pesto sauce (OR: 2.8; 95% CI: 1.3–5.8) remained as independent risk factors for disease in the multivariate model (Table 3).

Cluster in Sarpsborg

Four cases were reported from Sarpsborg (Figure 1). The first case sought medical attention and was tested for gastrointestinal pathogenic bacteria on 11 October and the last one on 25 October (Figure 2). The median age was 46.5 years (range: 45–64 years; Table 1). Three of the cases had eaten food containing fresh basil at the same restaurant in Sarpsborg ('restaurant Y') during the week before getting sick. The fourth case had not eaten in restaurant Y but lived with one of the other cases, suggesting person to person transmission. None of them had any link to the cases reported in Tromsø. None of these patients were hospitalised.

On 20 October, the NIPH posted an enquiry on the Epidemic Intelligence Information System (EPIS) hosted by the European Centre for Disease Prevention and Control to enquire if other European countries had reported clusters of *S. sonnei* infection. No information of any concomitant increase of cases of *S. sonnei* infections in other countries was received.

Microbiological investigation

The NRL received 48 *S. sonnei* isolates from stool samples during October 2011. Of these, 46 were confirmed by microbiological characteristics to be part of the outbreak. The MLVA-profiles of these isolates were identical (44 isolates) or with one locus difference (2 isolates). This MLVA-profile differed to a great extent from earlier profiles available in the NRL database including approximately 600 isolates and 405 distinct profiles.

The Norwegian Veterinary Institute analysed 20 food specimens from Tromsø and Sarpsborg consisting of diverse vegetables, fresh herb spices, fruits, nuts, herb dressings (including basil for pesto) and spiced butter. All food specimens analysed were negative for *Shigella* spp., but harboured high *Enterobacteriaceae* counts with relatively low levels of thermotolerant coliforms. One basil pesto product in particular originating from Delicatessen X had high levels of both *Enterobacteriaceae* and thermotolerant coliform counts.

Trace-back investigation

The Norwegian Food Safety Authority identified that the same supplier had provided fresh basil both to Delicatessen X in Tromsø and to restaurant Y in

Table 2

Univariate analysis of foods to which probable cases of *Shigella sonnei* infection (n=11) were exposed at a banquet in Tromsø, Norway, 1–2 October 2011

Food items	Exposed			Non exposed			RR	95% CI	Probable cases exposed (%)
	Probable cases	Total	AR	Probable cases	Total	AR			
Fish soup	10	21	47.6	1	20	5.0	9.5	(1.3–67.8)	90.9
Waldorf salad	8	16	50.0	1	18	5.6	8.9	(1.3–64.3)	72.7
Basil pesto sauce	7	9	77.8	4	28	14.3	5.4	(2.1–14.4)	63.6
Roast beef	7	14	50.0	2	19	10.5	4.8	(1.2–19.5)	63.6
Herb dressing	5	9	55.6	3	23	13.0	4.3	(1.3–14.2)	45.5
Mustard sauce	5	8	62.5	4	25	16.0	3.9	(1.4–11.1)	45.5
Mousse	6	9	66.7	4	23	17.4	3.8	(1.4–11.1)	54.5
Banana cake	6	9	66.7	5	27	18.5	3.6	(1.4–9.)	54.5
Raspberry sauce	6	10	60	4	24	16.7	3.6	(1.3–10.1)	54.5
Aioli sauce	6	11	54.5	4	23	17.4	3.1	(1.1–8.9)	54.5

AR: attack rate; CI: confidence interval, RR: relative risk.

For each food exposure, there were between one and 10 attendees missing a response because they did not recall having consumed or not a given food item.

Sarpsborg. The fresh basil was imported to Norway from Israel via the Netherlands. The Norwegian importer had received the basil on 25 September and delivered it to Tromsø on 27 September and to the restaurant in Sarpsborg on 27, 30 September and 4 October. No basil from the specific batch was left on the Norwegian market at the time when it was identified as the likely source of the outbreak. No further cases have been reported since 25 October.

On 11 November, following the identification of the implicated batch, the Norwegian Food Safety Authority generated a Rapid Alert System Food and Feed (RASFF) message about the basil, alerting other European countries.

Discussion

The results of the trawling interviews with the laboratory-confirmed cases in Tromsø made us hypothesise that an ingredient used in pesto served in Delicatessen X could be the source of the outbreak. The results of the cohort study among the banquet attendees reinforced this hypothesis: eating basil pesto was an independent risk factor for disease and had the highest attack rate among exposed. In addition, basil pesto was the only common food item that had been consumed by the other laboratory-confirmed cases occurring in Tromsø and Sarpsborg. The findings from the cluster investigation in Sarpsborg strongly supported fresh basil as the vehicle ingredient of *S. sonnei*. The role of the other food item highlighted in the cohort study, fish soup, remains unclear. We considered whether an ingredient of the basil pesto could also be part of the fish soup. The hypothesis was rejected as since the soup was not made by Delicatessen X, no common ingredients were used. None of the ingredients used in the soup had been eaten by the other laboratory-confirmed cases in Tromsø. The role of the food handlers in a potential cross contamination of the two food items remains unclear.

This outbreak, with 46 laboratory-confirmed cases, is the second largest shigellosis outbreak reported by 2013 in Norway [15]. A larger *S. sonnei* infection outbreak occurred during 1994 and affected several countries in Europe, including Norway, Sweden and the United Kingdom. In the 1994 outbreak, there were 110 laboratory-confirmed cases within Norway and investigations traced it to imported iceberg lettuce [8].

Table 3

Independent risk factors determined by multivariate analysis for probable *Shigella sonnei* infections at a banquet in Tromsø, Norway, 1–2 October 2011

Food items	OR (95% CI)	P value
Fish soup	8.2 (1.1–61.1)	0.04
Basil pesto sauce	2.8 (1.3–5.8)	0.01

OR: odds ratio; CI: confidence interval.

Several shigellosis outbreaks reported in Scandinavian countries have been associated with imported fruits or vegetables consumed raw or minimally-processed [7,16]. These food items might become contaminated during preparation by infected food handlers or during production by irrigation water contaminated with sewage [2]. The low infective dose and the considerable amount of fresh basil as an ingredient in pesto may have contributed to the large number of people becoming sick after eating basil pesto from delicatessen X in Tromsø, despite the growth-inhibitory effect of fresh herbs like basil or thyme on *S. sonnei* reported by some studies [17]. In this outbreak few affected individuals were admitted to hospital and no patients reported serious extra-intestinal symptoms. A noteworthy high C-reactive protein in affected patients has also been reported in previous studies of shigellosis [18].

Despite the epidemiological evidence which seemed to conclusively identify basil as the likely source of the outbreak, none of the specimens were positive for *Shigella*. Detection of *Shigella* spp. in food items is difficult and no reliable method is available. High levels of both *Enterobacteriaceae* and thermotolerant coliform counts were obtained from a suspected pesto product. This indicates faecal contamination and makes contamination also by *Shigella* more likely.

The Norwegian importer decided to temporarily stop importing basil from the exporter upon the identification of the batch. The exporter went bankrupt, so no decision on when to resume importation was necessary. It is unclear at which point in the process of cultivation, production and importation of the basil the *S. sonnei* contamination may have occurred. Currently, basil from certain third countries outside EU/European Free Trade Association (EFTA), as Israel, are not included in the European Commission regulation (EC) No 669/2009 list of certain feed and food of non-animal origin subjected to increased level of official controls on imports. As a result of this investigation, the Norwegian Food Safety Authority, together with NIPH, planned to develop recommendations for food providers on how to handle fresh plant produce prior to consumption.

The multidisplinary collaboration during this investigation helped to identify and find the source of this outbreak of *S. sonnei* infection in Norway: The routine genotyping of all isolates of enteropathogenic bacteria in Norway was crucial to determine that the two clusters happening in two regions of the country were part of the same outbreak. The epidemiological and product trace-back investigations pointed to imported fresh basil as likely causing the outbreak.

Acknowledgements

We would like to acknowledge the following colleagues for their contribution to this outbreak investigation: Alicia Barrasa and Yvan Hutin from the European Programme for Intervention Epidemiology training and Berit Tafjord Heier, Emily Macdonald, Heidi Lange and Kirsten Konsmo from the Norwegian Institute of Public Health. Parts of this work were presented as a poster at the Epidemic intelligence Service (EIS) Conference. Atlanta 22-26 April 2012.

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