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# NATIONWIDE OUTBREAK OF STEC O157 INFECTION IN THE NETHERLANDS, DECEMBER 2008-JANUARY 2009: CONTINUOUS RISK OF CONSUMING RAW BEEF PRODUCTS

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The Netherlands experienced a nationwide outbreak of Shiga toxin-producing *Escherichia coli* (STEC) O157 with onset of symptoms from the end of December 2008 until the end of January 2009. A total of 20 laboratory-confirmed cases were linked to the outbreak strain, serotype O157: H-, stx1, stx2, eae and e-hly positive. The investigation into the source of this outbreak is still ongoing, but evidence so far suggests that infection occurred as a result of consuming contaminated raw meat (steak tartare).

### Introduction

Shiga toxin-producing *Escherichia coli* (STEC) can cause bloody diarrhoea which progresses to a life-threatening condition known as the haemolytic uraemic syndrome (HUS) in 2-15% of the cases, particularly children [1,2]. STEC O157 is the serogroup most commonly identified in outbreaks.

STEC is found in the guts of many animals, but ruminants and cattle in particular are believed to be the main reservoir of human pathogenic STEC. Although infection can occur through direct contact with animals, most human infections are probably foodborne; water or food products (undercooked beef, raw milk, vegetables such as lettuce and alfalfa) contaminated with manure have often been linked to common source outbreaks. Person-to-person transmission can also occur [3,4].

An enhanced laboratory-based surveillance for STEC O157 was implemented in the Netherlands in 1999 and STEC non-O157 serogroups were added to this surveillance in 2007 [5]. Since 1999, STEC has been notifiable by law. All STEC-positive isolates identified in laboratories across the country are sent to the National Institute of Public Health and the Environment (RIVM). Since the start of the surveillance the number of STEC O157 infections reported annually ranged from 35 to 57 between 1999 and 2006, increased to 83 cases in 2007 as a result of a national outbreak [6], and dropped to 46 sporadic cases in 2008.

In week 4 of 2009, an increase to nearly half the predicted annual number of STEC O157 cases (n=20) was noted thanks

to the intensive surveillance system. This prompted a further epidemiological and microbiological investigation, the results of which are presented here.

### Methods

An outbreak investigation was initiated on 29 January 2009 in response to laboratory confirmation of a nationwide outbreak of STEC O157. An outbreak case was defined as a person diagnosed with a laboratory-confirmed STEC O157 infection since 10 December 2008 and a pulsed-field gel electrophoresis (PFGE) profile belonging to the outbreak cluster. Municipal Health Authorities in the Netherlands routinely follow up laboratory-confirmed STEC cases using a standardised questionnaire to collect information on clinical symptoms and exposure to possible risk factors in the week preceding onset of illness. Due to the dispersed distribution of cases within the Netherlands suspicion was raised that the cause could be a common food source or supplier; Municipal Health Authorities were requested to pay particular attention to the completeness of responses to questions pertaining to food history and location of purchase in their follow up of laboratory-confirmed cases.

All STEC positive isolates sent in to the RIVM are tested for genes encoding Shiga toxin type 1 and type 2 (*stx*<sub>1</sub> and *stx*<sub>2</sub>), the *E. coli* attaching-and-effacing gene (*eae*) and the enterohaemolysin encoding EHEC-*hly* gene (*e-hly*). DNA fingerprints are generated by PFGE, using *Xba*I as the restriction enzyme. The fingerprints are processed with BioNumerics® (Applied Maths, Kortrijk, Belgium; Dendrogram type=UPGMA, Similarity coefficient=Dice) [4].

### Statistical analysis

Analysis of food exposures was conducted using a case-case study design to compare laboratory-confirmed STEC O157 outbreak cases with sporadic cases of STEC O157 reported in the enhanced surveillance in 2008. Food items that were reported to have been definitely or possibly consumed were compared with items that were reported to have not been consumed. An attack rate and odds ratio for each food item was calculated using STATA 10. Individuals

who did not provide information on a food item were excluded from analysis of this particular food. Three possible secondary cases, defined as members of the same family as a case and with onset of symptoms later than that of the first family member, were also excluded from the analysis.

#### Food tracing

A trace back of suspected food items was initiated. The Food and Consumer Products Safety Authority (VWA) collected samples of any available left-over meat products from patients' homes for testing for STEC O157. The VWA also investigated the supermarkets and producers of various meat products mentioned by the cases.

#### International cooperation

The Netherlands is member of the European food and waterborne diseases and zoonoses surveillance network (formerly ENTERNET) administered by the European Centre for Disease Prevention and Control, which covers, amongst others, STEC infections. Using this network, in week 6 a request was made to all member countries to provide details of any occurrences of STEC O157 with a similar PFGE pattern.

#### Results

Between 27 December 2008 and 22 January 2009, 20 cases of STEC O157 (including three secondary cases) were attributed to the outbreak strain in the Netherlands (Figure 1). One additional STEC O157 case, with symptom onset on 13 December 2008, was possibly associated with the outbreak strain in accordance with the PFGE case definition and three isolates are pending PFGE typing to determine whether they are related to the outbreak.

Cases were spread throughout the Netherlands and were aged between 6 and 76 years of age (median age 41), with an equal number of males and females.

The outbreak strain was characterised as serotype O157:H-, and stx1, stx2, eae and e-hly positive and, all isolates, with the exception of one, were sorbitol-nonfermenting (Figure 2). This exact PFGE pattern has been seen on only two occasions in the Netherlands, both in 2005.

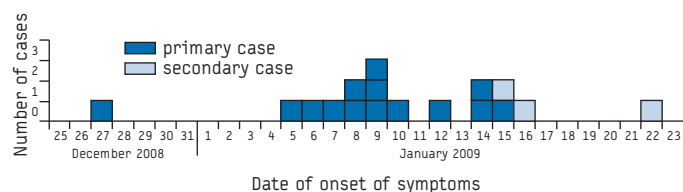
Sixteen of the 20 outbreak-related cases (80%) completed the questionnaire, three of which were secondary cases. Seven cases were hospitalised and none developed HUS. The questionnaire was

also returned by 36 non-outbreak cases of STEC O157 with onset of symptoms in 2008. These cases represented 78% of the total

**FIGURE 2**  
Pulsed-field gel electrophoresis (PFGE) pattern of the outbreak strain (middle three lanes) in the national outbreak of Shiga toxin-producing *Escherichia coli* (STEC) O157 in the Netherlands, December 2008 - January 2009



**FIGURE 1**  
Distribution of confirmed cases of the outbreak strain of Shiga toxin-producing *Escherichia coli* (STEC) O157 in the Netherlands, December 2008 - January 2009, by date of onset of symptoms (n=17)\*



\* Date of onset was unknown for three of the twenty outbreak-related cases

number of STEC O157 cases reported in 2008, ranged in age from 1 to 65 years and 67% were women. Two food items were frequently reported to have been consumed by outbreak cases: minced meat and steak tartare. There appears to be a link between consumption of steak tartare and STEC O157 infection: 83% of the 13 primary outbreak cases who provided the relevant information reported eating steak tartare in the week before illness compared to 18% of the sporadic 2008 cases (OR 16.3;  $p < 0.001$ , Table).

Many of the cases purchased food at more than one supermarket or store, and the precise location where individual items were purchased was not recorded. The supermarket or butcher where the implicated steak tartare was bought is therefore unknown.

### Results of food tracing

All left over food samples collected by the VWA tested negative for STEC O157. However, the food trace-back investigation is still ongoing and it is possible that the STEC cases pending PFGE results may be linked to the outbreak strain. Even if the contaminated batch of meat has been long-since consumed it is still worthwhile to further investigate the production flow in supermarkets, producers of steak tartare and possibly also slaughterhouses to try and gain information to support the epidemiological link.

### International response

Nine countries responded to the information request (Belgium, Germany, Finland, Ireland, Norway, England and Wales, Scotland, United States and Denmark), none of which reported current STEC O157 cases with PFGE profiles related to the outbreak strain. Finland reported two strains isolated in 1998 and 2004 with identical PFGE patterns, both patients had reported travel to Turkey.

### Conclusions

This is not the first time that a nationwide outbreak of STEC O157 has occurred in the Netherlands. In 2005, an outbreak was linked to steak tartare [3] and in 2007 an outbreak was associated with lettuce [4]. Based on the case-case study it seems probable that the outbreak reported here was also caused by steak tartare; steak tartare consumption was strongly and significantly associated with being an outbreak case and could explain the majority of the cases. The age distribution of cases seems also consistent with these findings; 28% of the non-outbreak cases of STEC O157 infection in 2008 were five years old or younger but the youngest outbreak-related patient was six years old which is not surprising as very young children are less likely to eat raw meat products.

TABLE

#### Risk factors for outbreak of Shiga toxin-producing *Escherichia coli* (STEC) O157 in the Netherlands, December 2008 – January 2009

	Outbreak case (n=13)	Non-outbreak case (n=36)	Univariate odds ratio	
Food item	n (%)	n (%)	OR (95%CI)	p-value
Minced meat	10 (77)	21 (60)	2.2 (0.4-14.5)	0.23
Steak tartare	10 (83)	6 (18)	16.3 (3.2-238.6)	<0.001

Note: Percentages were calculated taking as denominator the number of persons who provided the relevant answer

The incubation period of STEC O157 is generally considered to be 1-10 days. Thirteen of the 14 primary cases with known date of symptom onset became ill within 11 days of each other. Raw meat can become contaminated during slaughter, and by cutting and mixing the meat a point source contamination can result in the contamination of a large batch of meat. Hygienic slaughter processes are imperative but contamination of carcasses cannot be completely avoided. This outbreak is another sign that despite control measures and legislation, raw meat products continue to pose a risk for the health of the general population. Raising consumer awareness in relation to consumption of raw meat is still needed [1,7,8].

Tracing of meat products continues to be a difficult task because insufficient detail is collected in the routine questionnaires about the precise type of meat, such as whether it is beef or veal, pre-packed or fresh from a butcher. Although questionnaire data is very useful, in our investigation several cases mentioned shopping at more than one supermarket chain without distinguishing which items were purchased where. This made it difficult to trace back the place of production and purchase of the implicated steak tartare. Obtaining supermarket receipts from cases could assist with the trace back, particularly in investigations in which one supermarket chain is frequented or when it is unclear what products were purchased where. This could improve the efficiency of the food trace back, but it is also time-consuming when more than one supermarket is involved and there is no protocol in place. It would also be useful to collect detailed information about steak tartare in the routine questionnaire. It is also apparent that we still do not have a good insight into the production chain of steak tartare, despite two large outbreaks in recent years.

Even though the current outbreak was confined to the Netherlands, international trade in meat and vegetable products makes it important to raise the alert internationally and rapidly and accurately trace suspected food items. However, effective prevention of future outbreaks caused by consumption of steak tartare may be very difficult.

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