Outbreak report

Outbreak of *Salmonella* Enteritidis phage type 14b, May-September 2014
About Public Health England

Public Health England exists to protect and improve the nation's health and wellbeing, and reduce health inequalities. It does this through world-class science, knowledge and intelligence, advocacy, partnerships and the delivery of specialist public health services. PHE is an operationally autonomous executive agency of the Department of Health.

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Abbreviations and glossary

CCDC    Consultant in Communicable Disease Control
EHO     Environmental Health Officer
EPIS    Epidemic Intelligence Information System
FBO     Food business operator
FSA     Food Standards Agency
FWE     Food Water and Environment
GEZI    Department of Gastrointestinal, Emerging & Zoonotic Infections
GP      General Practitioner
HPT     Health Protection Team
IMT     Incident management team
MLVA    Multilocus Variable-Number Tandem-Repeat Analysis
OCT     Outbreak Control Team
PCR     Polymerase chain reaction
PHE     Public Health England
PHEC    Public Health England Centre
PT14b   Phage type 14b
SNP     Single nucleotide polymorphism
RASFF   Rapid Alert System for Food and Feed
WGS     Whole genome sequencing
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Executive Summary

Background
At the beginning of June 2014, Public Health England (PHE) was alerted to an outbreak of *S. Enteritidis* phage type (PT) 14b in a hospital in central England. Following this, outbreaks of *S. Enteritidis* PT14b cases were detected in the North West and South of England. A national Outbreak Control Team (OCT) was formed to co-ordinate a multi-disciplinary investigation to understand the source of this outbreak and enable suitable control measures to be put in place to prevent further cases.

Investigations
The epidemiology of cases was described at the national level. Outbreaks associated with specific premises were investigated locally. Microbiological investigations, including whole genome sequencing (WGS), were undertaken for isolates from cases and food samples from the UK and abroad. Food chain investigations examined the supply chain for egg and chicken products eaten by each case. Genetic sequencing investigated the genetic relatedness of isolates from human and food samples.

Findings
In total, 287 cases met the definition; ages ranged from <1 to 92 years (median 29); 151 cases (53%) were male. In total, 198 of 287 (69%) cases could be plausibly linked to eggs supplied by one company, Company X. The WGS results showed that the 369 clinical and environmental samples from England, Germany, France, Luxembourg and Austria formed a tight outbreak on the *Salmonella* Enteritidis phylogeny.

Conclusion
This report describes the national investigation of an outbreak of gastrointestinal illness caused by *Salmonella* Enteritidis PT14b. This report presents whole genome sequencing data which provide a clear link between isolates from cases, eggs and environmental samples from premises associated with outbreaks of cases. This, along with the egg supply food chain information and information from investigations in other European countries, provides compelling evidence to support the hypothesis that this outbreak was associated with eggs from Company X.

Recommendations
- We recommend that the FSA work with local authorities and food business operators (FBOs) to ensure that they are fully compliant with the traceability requirements of Article 3 of Regulation (EC) 931/2011.
- We recommend that Members States support measures to create a framework to ensure that public health control measures are enhanced by the exchange of pathogen sequencing information.
Background

Public Health England undertakes routine surveillance which aims to detect outbreaks of infectious disease so that further cases can be prevented by implementing timely and effective control measures. This report describes the national response to a Salmonella outbreak.

Salmonella is a common cause of bacterial gastroenteritis in the United Kingdom. [1] Salmonella bacteria are widely distributed in domestic and wild animals. Salmonellosis in humans is generally contracted through the consumption of contaminated food items, though person-to-person transmission through the faecal-oral route can also occur.

The adoption of vaccination and other measures in management of poultry production has led to a reduction in the number of Salmonella enterica serovar Enteritidis infections in the United Kingdom.[2] Despite this reduction, there have been several outbreaks of Salmonella Enteritidis phage type 14b (PT14b) in the United Kingdom which have been associated with imported chicken eggs.[3,4]

Coordination of the response

How the incident came to light
At the beginning of June 2014, Public Health England (PHE) was alerted to an outbreak of S. Enteritidis PT14b in a hospital in central England. Following this, outbreaks of S. Enteritidis PT14b cases were detected in the North West and South of England.

In addition, PHE was alerted through the Epidemic Intelligence Information System (EPIS) to six Salmonella outbreaks in France associated with eggs from Germany and an Austrian Salmonella outbreak, both countries reporting matching MLVA (Multilocus Variable-Number Tandem-Repeat Analysis) profiles.

Following the detection of an exceedance in the cumulative number of S. Enteritidis PT14b cases in England and Wales, PHE launched a national investigation; the first national outbreak control team meeting was held on 6 August 2014.
### Chronology of events (2014)

<table>
<thead>
<tr>
<th>Date</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>09 July</td>
<td>RASFF notification issued by France regarding a <em>Salmonella Enteritidis</em> outbreak suspected to be caused by eggs from Germany (with distribution to France and Hungary)*</td>
</tr>
<tr>
<td>17 July</td>
<td>Last delivery of Company X eggs from German sites</td>
</tr>
<tr>
<td>31 July</td>
<td>RASFF notification issued by Austria regarding a <em>Salmonella Enteritidis</em> outbreak suspected to be caused by eggs from Germany (with distribution to Austria, Croatia, Czech Republic, and Slovakia)*</td>
</tr>
<tr>
<td>01 August</td>
<td>Second RASFF notification issued by France regarding a <em>Salmonella Enteritidis</em> outbreak suspected to be caused by eggs from Germany (with distribution to Austria, Czech Republic, France and Germany)*</td>
</tr>
<tr>
<td>04 August</td>
<td>MLVA results received from France</td>
</tr>
<tr>
<td>06 August</td>
<td>First OCT meeting</td>
</tr>
<tr>
<td>12 August</td>
<td>OCT meeting; outbreak declared as PHE level 3</td>
</tr>
<tr>
<td>13 August</td>
<td>PHE briefing note issued</td>
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<tr>
<td>15 August</td>
<td>PHE issued proactive press release</td>
</tr>
<tr>
<td>20 August</td>
<td>OCT meeting</td>
</tr>
<tr>
<td>27 August</td>
<td>OCT meeting</td>
</tr>
<tr>
<td>01 September</td>
<td>Last delivery of Company X eggs from the Czech site</td>
</tr>
<tr>
<td>03 September</td>
<td>OCT meeting</td>
</tr>
<tr>
<td>10 September</td>
<td>OCT meeting</td>
</tr>
<tr>
<td>17 September</td>
<td>OCT meeting</td>
</tr>
<tr>
<td>24 September</td>
<td>OCT meeting</td>
</tr>
<tr>
<td>01 October</td>
<td>OCT meeting</td>
</tr>
<tr>
<td>22 October</td>
<td>OCT meeting; outbreak closed as cases returned to expected levels</td>
</tr>
<tr>
<td>12 December</td>
<td>OCT debriefing</td>
</tr>
</tbody>
</table>

*Initial RASFF notifications did not indicate that potentially affected eggs had been distributed to the UK*
Investigations

Epidemiological

Case definition
Outbreak cases were defined as any primary indigenous case of *Salmonella* Enteritidis PT 14b, resident in England and Wales, reported after 1 June 2014, with a MLVA profile of 2-11-9-7-4-3-2-8-9, or a single locus variant thereof (the MLVA outbreak profile).

Case finding
Case ascertainment was from statutory notifications of *Salmonella* infection. *Salmonella* Reference Service conducted phage typing and MLVA typing to distinguish those infections which met the case definition.

Microbiological typing
*S. Enteritidis* strains conformed to the recognized pattern for phage type 14b. Isolates were further characterised by MLVA typing and whole genome sequencing (WGS). MLVA comprises of a multiplex PCR which specifically amplifies repeated sequences from nine different loci followed by fragment size analysis using multicolour capillary electrophoresis. Sequencing was carried out by the PHE Genome Sequencing Unit using Nextera library preparation and the Illumina HiSeq 2500 in fast run mode according to manufacturers’ instructions.

Food chain investigation
Cases were interviewed using local questionnaires to ascertain foods eaten in the five days before onset of symptoms. The process of obtaining this information was led by Public Health England and local authorities. Shops, restaurants and other food outlets reported by cases were identified and food chain investigations were led by the Food Standards Agency.

Local investigations
Several local outbreaks of *S. Enteritidis* PT 14b were identified to be associated with particular premises. These were subject to local investigation and outbreak control activities. Descriptive epidemiology, microbiological and environmental investigations were undertaken. Food chain information obtained from cases and premises was used to inform the national investigation.
International investigations
As international communications had identified that cases had also occurred in other European countries, WGS was undertaken on isolates from outside the UK. This included four isolates from France; two human cases and two from eggs originating from Company X. Two human isolates were received from Luxembourg, one from 2010. Six isolates were received from Austria, all from cases. Fourteen isolates were received from Germany; five from cases, one from a cake and eight from eggs from Company X (six from one company X site, two from another Company X site).

Results
Epidemiological
In total, 287 cases met the definition; ages ranged from <1 to 92 years (median 29) 151 (53%) were male. A population pyramid detailing case demographics is shown in Figure 1. The spatial distribution of cases is shown in Figure 2. Due to the number of cases associated with particular premises, the cases show a high level of geographic clustering.

Figure 1: Age and sex of cases (n=287)

Symptom onset dates ranged from 25 May 2014 to 7 September 2014. The week of symptom onset is shown in Figure 3; this also includes information on the residence of cases.
Figure 2: Location of cases by postcode of residence (n=287)

Figure 3: Epidemic curve showing onset date by ONS week (n=284)
Seventy-eight (27%) cases were reported to have been hospitalised (of whom 61 were not thought to have acquired their infection while in hospital).

**Local investigations**

**Hospital A outbreak**

Between 25 May and 18 June 2014, 32 cases (patients, staff and visitors) were linked to a single hospital in central England, of whom 17 had spent the whole incubation period in the hospital. *Salmonella* infection was considered to be a contributory factor in the cause of death for one patient. Following epidemiological and environmental investigations, *Salmonella* was isolated from a catering trolley on one of the wards.

**Restaurant A outbreak**

Ninety-nine cases were linked to restaurant A in southern England. Cases reported eating at the restaurant between 11 July and the closure of the restaurant on 24 July to improve food hygiene practices.
Restaurant B outbreak
Between 8 and 19 July 2014, 31 cases were linked to restaurant B in North West England. *Salmonella* was isolated from food samples (cooked chicken and pork) and an environmental sample (a cleaning cloth) taken at the premises.

Restaurant C outbreak
Fourteen cases were linked to restaurant C in central England. Cases reported symptom onsets between 24 July and 6 August 2014. The restaurant closed voluntarily between 7 and 13 August 2014. One further case occurred after this on 1 September. A dishcloth, a swab from a vegetable preparation sink and a sample of egg-containing vegetarian noodles all tested positive for *Salmonella*.

The food and environmental samples from outbreaks at hospital A, restaurants B and C which were positive for *Salmonella* were typed as S. Enteritidis PT14b, with those from hospital A and restaurants B and C having the same MLVA profile as cases. Six eggs of Spanish origin were sampled from restaurant C on 12 August 2014; all tested negative for *Salmonella*.

**Food chain investigation**
In addition to the information received through EPIS, Rapid Alert System for Food and Feed (RASFF) notifications were issued on 09 July 2014 (France), 31 July 2014 (Austria) and 1 August 2014 (France), linking S. Enteritidis outbreaks to chicken eggs from Germany. Subsequent updates to the RASFF notifications indicated that the outbreaks were caused by *Salmonella* Enteritidis PT14b. Because of this, food chain investigations in England traced supplies of chicken and chicken eggs to their source where possible.

In total, 198 of 287 (69%) cases could be plausibly linked to eggs supplied by one company, Company X (see Figure 4). Thirty-two cases ate at premises where eggs with a Company X egg stamp number were observed; 166 cases were linked to premises that were plausibly supplied by Company X at the time of exposure, according to information from suppliers. Restaurants A, B and C were all supplied with eggs originating from Company X, as was one outlet at hospital A (although most cases did not report eating food from this outlet).

Company X has four sites, of which three are in Germany and one is in the Czech Republic; all are operationally independent. All four sites use young chickens (pullets) from two sites: one in Germany and one in the Czech Republic. The last delivery to the UK from all three German sites was on the 17 July 2014; the last delivery of eggs from the Czech Republic site was on 1 September 2014.
Control measures
Environmental health officers investigated premises associated with outbreaks to ensure compliance with food hygiene guidelines. The available evidence indicated that potentially affected eggs had been supplied to catering establishments in the UK. On 22 August 2014 the FSA sent letters to local authorities to ask them to contact catering premises in their area and reiterate FSA advice on how to cook and prepare eggs safely. Additionally, the letters asked local authorities to look in catering establishments for eggs with the three egg stamp numbers relating to Company X’s German premises. No reports of finding these eggs were received. On 22 August 2014, caterers were also reminded of the guidelines for the safe handling of eggs via the FSA website. In addition, enhanced infection control procedures were introduced in hospital A to reduce the risk of person to person spread.
Microbiological Results
The human isolates from France, Austria, Germany and Luxembourg all had the outbreak MLVA profile. The isolates from Company X eggs shared the outbreak MLVA profile. The WGS results showed that the 369 clinical and environmental samples from England, Germany, France, Luxembourg and Austria clustered...
phylogenetically on the *Salmonella* Enteritidis phylogeny (Figure 5). Within the outbreak cluster the minimum SNP distance between strains was 0 and the maximum was 23 SNPs. Within the hospital outbreak A and the restaurant outbreaks A, B and C the mode SNP distance was 0 SNPs with no clinical isolate differing by greater than 2 SNPs. Implicated environmental isolates were either identical or a single SNP away from a clinical isolate. Clinical isolates from the rest of Europe clustered within clinical isolates from England as did all eight isolates from eggs from Company X.
Figure 4: Phylogeny of *Salmonella* Enteritidis isolates, including representative isolates from England and Wales (n=484), sequenced between January 2012 and September 2014 together with outbreak isolates (n=369)
Communications
Public Health England issued a media release on 15 August. The statement was issued on behalf of the OCT and provided information on the investigation. National and local media interest was received. Dr Paul Cleary undertook radio interviews with the BBC.

Discussion and conclusions
This outbreak demonstrated the importance of MLVA which was used to identifying this multi-country Salmonella outbreak, and the use of whole genome sequencing which provided further confirmation. WGS allows improved discrimination between isolates, and adds a new dimension to descriptive epidemiology in the form of phylogenetic relationships. WGS has previously been used to confirm a multi-country Salmonella outbreak in Europe [6], but the outbreak reported here is the first time it has been used in “near real-time” to define a multi-country Salmonella outbreak and inform public health control measures.

Limitations
Despite testing, no eggs sampled in the UK as part of the outbreak response were positive for Salmonella; this most likely reflects time elapsed between egg consumption, symptom onset, phage typing, food history taking and egg sampling. The delay between egg consumption and sampling is usually greater than the shelf life of eggs which is typically 26 days (Mark Jones, AHVLA, personal communication, 24 September 2014), making it inherently unlikely that eggs identified at catering premises during this outbreak investigation were the ones consumed by cases.

Conclusion
This report describes the national investigation of an outbreak of gastrointestinal illness caused by Salmonella Enteritidis PT14b. This report presents whole genome sequencing data which provide a clear link between isolates from cases, eggs and environmental samples from premises associated with outbreaks of cases. This, along with the egg supply network information and information from investigations in other European countries, provides compelling evidence to support the hypothesis that this outbreak was associated with eggs from Company X.

The OCT concludes that the most plausible explanation is that this outbreak was caused by the consumption of items contaminated by Salmonella from eggs produced by Company X.
Recommendations

We present genetic and food chain information which support the hypothesis that this multi-national *Salmonella* Enteritidis PT14b outbreak was associated with eggs from Germany. This investigation demonstrates the importance of European cooperation in complex food supply networks. Information, both official and informal, from other European countries was important in both detecting the outbreak and ensuring that public health actions in the UK were as timely as possible.

Current EU regulations [5] provide extensively for traceability of products of animal origin. Article 3 requires FBOs to ensure that particular information concerning consignments of food of animal origin is made available to the FBO to whom the food is supplied and, upon request, to the competent authority. This includes information on the volume or quantity of the food, a reference identifying the lot, batch or consignment, as appropriate, an accurate description of the food, the date of dispatch and the name and address of the FBO from which and to whom the food has been dispatched.

However, it was the experience of investigators in this outbreak that for many FBOs, this information was very difficult to obtain. Specifically, lot/batch/consignment identifiers were frequently not recorded by FBOs, or done so in a way that made it very challenging to effectively trace the movement of the eggs associated with this outbreak.

- We therefore recommend that the FSA work with local authorities and FBOs to ensure that they are fully compliant with the traceability requirements of Article 3 of Regulation (EC) 931/2011.

Being able to sequence isolates from German eggs made the genetic evidence linking this source to UK cases more compelling.

- We therefore recommend that Members States support measures to create a framework to ensure that public health control measures are enhanced by the exchange of pathogen sequencing information.
References


