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Outbreak of invasive pneumococcal disease at a Belfast shipyard in men exposed to welding fumes, Northern Ireland, April–May 2015: preliminary report

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We report an outbreak of four confirmed cases of invasive pneumococcal disease (IPD) in individuals occupationally exposed to welding fumes, at a Belfast shipyard (Northern Ireland). All cases were hospitalised. A high-risk sub-group of 679 workers has been targeted for antibiotic prophylaxis and pneumococcal vaccination. Physicians and public health institutions outside Northern Ireland should be alert to individuals presenting with pneumonia or IPD and recent links to the shipyard, to facilitate early assessment and treatment.

Incident description

On 29 April 2015, two cases of invasive pneumococcal disease (IPD) with dates of onset of illness on 28 April were notified to the regional Health Protection Service, Public Health Agency (PHA) Northern Ireland. The only epidemiological link that could be identified was that they both worked on the refurbishment of an oil rig at a Belfast shipyard. PHA signposted the shipyard to the United Kingdom (UK) Health and Safety Executive (HSE) guidelines and reinforced the importance of personal protective equipment (PPE). The isolates were sent for typing to the Public Health England (PHE) Respiratory and vaccine preventable bacteria reference unit (RVPBRU). On 13 May, the PHA was notified of two further cases of IPD with onset dates on 6 May, in men who worked at the same shipyard.

The cases were in men aged in their 20s to 50s from the UK and two other European countries. All were from occupational groups potentially exposed to welding fumes. All presented with pneumonia, and were microbiologically confirmed by isolation of *Streptococcus pneumoniae* from blood cultures (three cases) or detection of urinary antigen (one case). All isolates were fully sensitive to amoxicillin and erythromycin. Typing of all three blood culture isolates revealed serotype 4; the urinary antigen positive sample could not be typed.

Shipyard workforce

Up to 1,500 individuals can be on site with a multi-national staffing component including for example Bulgarian, Lithuanian, Norwegian, Polish, Romanian and Russian nationalities (approximately one third of all staff). The workforce consists of core workers and temporary / agency staff who are employed by a range of contractors, themselves also based in different European countries. The obligation for occupational health provision lies with the contractors for their respective workers.

Epidemiological investigations

We are using a specific case definition for the purpose of the outbreak investigation and active case finding. A confirmed case is an individual who has worked at the Belfast shipyard since mid-January 2015 with a clinical diagnosis of IPD or pneumococcal pneumonia AND at least one of the following: pneumococcus isolated from normally sterile site (blood, cerebrospinal fluid (CSF), joint, peritoneum, pleural fluid or other, but not sites such as eye), pneumococcal DNA or antigen detected in fluid from a normally sterile site or pneumococcal antigen detected in urine.

We are collecting information on confirmed cases to include patient identifiers and demographic characteristics, risk factors for IPD, clinical details, microbiological investigations, vaccination status and onsite working patterns [1].

Retrospective case finding will be conducted through examination of laboratory-confirmed reports in people aged 18–64 years with specimen dates in 2015. Prospective case finding is undertaken through local, national and international alerting. Retrospective typing of isolates from IPD cases during 2015 will be carried out to determine circulating pneumococcal serotypes.

No family members were identified as close contacts in the two days before the onset of symptoms in the cases. Due to the complex working patterns of the cases, no close work contacts, as defined in the Interim UK guidelines for the public health management of clusters of serious pneumococcal disease in closed settings, could be identified [1].

A communication was issued through the European Union early warning and response system (EWRS) on the morning of 15 May to facilitate case finding across Europe.

Background

Streptococcus pneumoniae is a Gram-positive coccus known to colonise the nasopharyngeal tract and can cause a spectrum of disease such as otitis media, pneumonia and invasive pneumococcal disease [2]. The virulence of the organism is determined by the capsule and over 90 different capsular types (serotypes) have been identified [3]. Clusters of IPD have been reported from closed settings including long-term care facilities, hospitals and households [4]. A number of risk factors have been identified for IPD including extremes of age, certain co-morbidities, smoking and immunosuppression [5]. Welders have an increased risk of IPD, and although not fully understood, this may relate to components of the fumes serving as a nutrient to increase adherence of the pneumococci to the lung tissue or inhalation of the fumes causing damage to the lung's immune defences [5-7]. The UK HSE guidelines recommend that 23-valent pneumococcal polysaccharide vaccine 'should be considered for people whose work exposes them to frequent or continuous exposure to metal fume (e.g. welders), taking into account the exposure control measures in place' [8].

S. pneumoniae isolates from normally sterile sites are reported to the PHA through routine voluntary laboratory reporting arrangements. Based on laboratory reporting, the average background rate of IPD in Northern Ireland (population 1.82 x10⁶) was ca 6 cases per 100,000 population during 2010–2014, with higher rates in the very young and in those aged over 65 years. Not all isolates are routinely serotyped.

Control measures

A multidisciplinary outbreak control team (OCT) meeting was held on 14 May with a range of local stakeholders, shipyard management and PHE. It was agreed that as no close contacts of the cases could be identified, those groups most exposed to welding fumes would be offered antibiotic prophylaxis with azithromycin or amoxicillin and vaccination with 23-valent pneumococcal polysaccharide vaccine (PPV23) [1,7]. Azithromycin 500 mg once daily for three days was the preferred first-line antibiotic, with a view to optimising compliance. For those with contraindications to azithromycin, amoxicillin 500 mg twice daily for seven days was offered. PPV23 vaccine was used as the incident was taken as an opportunity to protect this high-risk

occupational group not only against serotype 4, but also against a wider range of serotypes which might be encountered in future [9].

Antibiotic prophylaxis and vaccination was offered to anyone who was considered at risk of contracting IPD, i.e. worked as a welder or in other occupations with prolonged exposure to metal fumes in enclosed spaces in the working environment. A total of 679 among the around 1,500 shipyard workers met this definition and attended the clinics for the intervention between 16 and 18 May. Only six of the attendees were aware that they had received the pneumococcal vaccine previously. A multi-disciplinary team of public health and clinical staff, including pharmacists, administrators, translators and shipyard staff delivered the response. Dedicated clinics operated for between eight and eleven hours each day at times appropriate to ensure day and night shift staff had the opportunity to attend.

Conclusion

This investigation is ongoing and we hope to complete this preliminary information that aims to support our active case finding, with a more complete account, at a later date. A letter has been issued in Northern Ireland to local general practitioners, emergency departments and clinicians to increase awareness of the incident and request that all cases of IPD in shipyard workers are notified to the PHA prospectively.

If public health institutions from other countries are aware of further cases of IPD in individuals who have been working at the Belfast shipyard, Northern Ireland, since the middle of January 2015, we would like to invite them to contact the PHA duty room at pha.dutyroom@hscni.net in order for us to gain a full overview of the number of cases in this outbreak.

Conflict of interest

None declared.

Authors' contributions

LJ led the overall response to the incident and AW is the Port Health lead. Epidemiological investigations were conducted by NI and LP. LD is the Assistant Director for Health Protection and chaired the outbreak control team meetings. Microbiological investigations were conducted by AL. The manuscript was drafted by LP. All authors commented and agreed on the final manuscript.

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A prolonged outbreak of invasive meningococcal disease in an extended Irish Traveller family across three Health Service Executive (HSE) areas in Ireland, 2010 to 2013

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Between March 2010 and November 2013 eight laboratory-confirmed cases of serogroup B, invasive meningococcal disease (IMD) were identified in an extended Irish Traveller family across three Health Service Executive (HSE) areas of Ireland. Cases were aged between 5 and 46 months, and were either a cousin or sibling of another case. All eight cases survived. Chemoprophylaxis was given to relevant nuclear family members and close contacts on each occasion, but failed to prevent further cases. *Neisseria meningitidis* isolates from six cases were highly related, belonging to the ST-41/44 clonal complex, and shared the *porA* designation 7-2,4. In November 2013, the outbreak control team recommended that directly observed ciprofloxacin chemoprophylaxis be administered simultaneously to the extended family, and that the four component meningococcal B (4CMenB) vaccine be administered to family members aged 2 months to 23 years inclusive and relevant close contacts of the eighth case. Subsequently these recommendations were implemented at three regional clinics. Additionally pharyngeal swabs (n=112) were collected to assess carriage rates of *N. meningitidis* in this extended family. Pharyngeal carriage of *N. meningitidis* was detected in 15 (13%) family members. From the epidemiological investigation and carriage study overcrowding was the most likely risk factor identified in this outbreak. To date, the combination of directly observed ciprofloxacin chemoprophylaxis and use of 4CMenB vaccine have controlled the outbreak with no further cases diagnosed.

Introduction

Invasive meningococcal disease (IMD) is a life-threatening infection caused by *Neisseria meningitidis*. Risk factors for IMD include close contact with a known case, crowded environments, recent upper respiratory tract

infection or influenza, cigarette smoking and immunological susceptibility [1].

Under Irish law, clinicians and clinical directors of laboratories must immediately notify suspected cases of IMD to the Medical Officer of Health (MOH) at the local Department of Public Health. Laboratories must also immediately notify IMD cases which meet the laboratory criteria for confirmed or probable disease in accordance with the case definition [2].

All notified cases of IMD are entered on to the Computerised Infectious Disease Reporting (CIDR) database which is hosted by the Health Protection Surveillance Centre (HPSC), Ireland's specialist agency for the surveillance of communicable diseases. CIDR is a shared national information system, developed to manage the surveillance and control of infectious diseases in Ireland [3].

In 2011, Ireland had the highest incidence of meningococcal disease in the European Union at 1.99 per 100,000 population [4]. However, this figure represents a decrease of 86% from 14.8 per 100,000 population reported in 1999. Irish Travellers are an indigenous minority in Ireland. Their lifestyle and culture, which may include a nomadic lifestyle, distinguishes them from the general population [5]. Irish Travellers experience social and cultural marginalisation in a similar manner to other indigenous minorities globally [6].

In Ireland, ethnic identification is not routinely collected on all individuals with notifiable infectious diseases. Consequently, assessing national rates of disease by ethnicity is not possible. However since 2004, when the CIDR system became operational, limited data on ethnicity is available. Based on such data, it is evident that while the incidence of IMD has decreased in the

FIGURE

Cases of invasive meningococcal disease in an outbreak affecting an extended Irish Traveller family, Ireland, 2010–2013 (n=8)



general population, a similar reduction has not been seen in the Irish Traveller population. In contrast, since 2004 there has been an increase in reported cases of IMD in this population group [7].

In 2013, 84% of meningococcal disease in Ireland was caused by *N. meningitidis* serogroup B [8]. While most cases of IMD occur sporadically, occasionally an unusual cluster or outbreak is reported. The National Guidelines on the Clinical and Public Health Management of Bacterial Meningitis defines an outbreak of meningococcal disease as ‘a minimum of four cases of definite meningococcal disease within a three-month interval, or 40/100,000 in any age group in a three month period, in a geographical area that makes epidemiological sense AND where available microbiological characterisation of the organism is the same’ [9].

We describe the identification of eight cases of IMD due to *N. meningitidis* serogroup B in an extended Irish Traveller family over a three and a half year period across three of eight public health areas of the Health Service Executive (HSE) in Ireland. All eight cases occurred in young children (≤ 46 months) who were siblings or first cousins of identified cases in the outbreak. The measures to prevent further cases are described. Although the cluster formed by the eight cases does not fulfil the criteria for the above outbreak definition, the term ‘outbreak’ is used in this report to reflect the high endemicity within this extended family.

Methods

Case definitions

In Ireland, case definitions are specified for the purpose of reporting of selected infectious diseases. Clinical symptoms and signs suggestive of IMD include meningeal signs, haemorrhagic (petechial or purpuric) rash and septic shock [10].

A confirmed case requires one of the following laboratory criteria:

- Isolation of *N. meningitidis* from a normally sterile site or from a haemorrhagic skin lesion.
- Detection of *N. meningitidis* nucleic acid from a normally sterile site or from a haemorrhagic skin lesion.
- Detection of *N. meningitidis* antigen in cerebrospinal fluid (CSF).

The case definition used in this outbreak was ‘A laboratory-confirmed case of IMD caused by *N. meningitidis* serogroup B in a person who is a member of this extended Irish Traveller family’.

Strain characterisation and antimicrobial susceptibility testing

Laboratories refer all confirmed meningococcal samples to the Irish Meningococcal and Meningitis Reference Laboratory (IMMRL) for further characterisation. Specimens from all eight cases reported here were processed in the IMMRL by polymerase chain reaction (PCR) testing [11,12]. Characterisation of *N. meningitidis* isolates using multilocus sequence typing (MLST) [13], *porA* [14] and *fetA* [15] variable region fine-typing was also undertaken at the Epidemiology and Molecular Biology Unit (EMBU), which is linked to the IMMRL.

Antimicrobial susceptibility testing to cefotaxime, ciprofloxacin, penicillin, rifampicin and sulfadiazine was performed using Etest methodology according to the manufacturer’s instructions (BioMérieux).

Control measures for cases of meningococcal disease

When a case of suspected or confirmed meningococcal disease is reported to the MOH by a clinician or laboratory the following public health actions are carried out in order to prevent secondary cases; contact is made by public health doctors with the clinician responsible for the care of the case to discuss the clinical situation, contact is made with the case or next of kin to provide information on the illness and to identify close contacts, as defined in detail in the national guidelines [9], who require prompt chemoprophylaxis and possibly vaccination. Evidence-based information is also disseminated to local general practitioners (GPs), family members and, where relevant, schools and pre-schools.

Carriage study

A carriage study was conducted to estimate the prevalence of *N. meningitidis* carriage in the extended Irish Traveller family in which outbreak cases had been identified. The objective was to see if this family had a higher rate of carriage than that seen in the general population which may have contributed to increased transmission to and increased disease in the young children. A posterior pharyngeal swab was collected from all family members who attended special outbreak control clinics. These swabs were anonymised but selected demographic data was collected including;

TABLE 1

Description of cases of invasive meningococcal disease in an outbreak affecting an extended Irish Traveller family, Ireland, 2010–2013 (n=8)

Case	A	B	C	D	E	F	G	H
HSE area	East	East	East	Mid-west	East	East	South-east	East
Date of diagnosis	09/03/2010	20/11/2010	08/03/2011	01/01/2012	20/03/2013	29/04/2013	12/06/2013	28/11/2013
Clinical symptoms, signs	Fever, purpuric rash, cough, vomiting	Fever, purpuric rash, listlessness	Bulging fontanel, cough, fever, lethargy, poor feeding, purpuric rash	Fever, non-blanching rash, vomiting	Disseminated intravascular coagulation (DIC), fever, non-blanching rash	Anorexia, fever, irritability, lethargy, non-blanching rash	Fever, irritability, non-blanching rash	Eye swelling, lethargy, limb swelling, not drinking
Pre-hospital antibiotics	Not known	Benzylpenicillin	Benzylpenicillin	Not known	Benzylpenicillin	No	No	No
Length of hospital stay	13 days	10 days	8 days	8 days	14 days	12 days	8 days	23 days
Number of close contacts	18	7	19	16	8	26	33	18
Chemoprophylaxis (number of close contacts)								
Rifampicin	17	7	18	16	8	25	31	18
Ceftriaxone	1	0	1	0	0	1	2	0

HSE: Health Service Executive.

Fever is defined as body temperature $\geq 37.5^{\circ}\text{C}$.

In some of cases, a previous case was a close contact. For example, case A was a close contact in case C. Case B was a close contact in case H, case F was a close contact in case E.

age, sex, housing type and size, number of people living in household, pregnancy, contact with a previous case and previous chemoprophylaxis.

Pharyngeal swabs were cultured for *N. meningitidis* and processed for analysis using a *ctrA;porA* duplex real-time *N. meningitidis*-specific PCR assay (data not shown) [12]. Characterisation of *N. meningitidis* isolates using serotyping, genogrouping, *porA/fetA* fine-typing, multilocus restriction typing (MLRT) and MLST was also undertaken [13–18]. Antimicrobial susceptibility testing was performed on isolates as above.

Results

Description of the outbreak

Between March 2010 and November 2013 eight cases met the case definition (Figure).

Cases are described from A to H in chronological order (Table 1). At the time of the third case's illness (case C), it was confirmed that this case's sibling (case A) and first cousin (case B) had been reported as having IMD the previous year (2010). In April 2013, following the identification of two additional family cases, one in March 2013 and the other in April 2013 an outbreak was reported based on the recognition of a cluster of cases over a prolonged period of time. This led to the further identification of affected individuals including the retrospective finding of a case in 2012 (Figure). All cases were less than four years-old (median age: 7.5 months; range: 5–46 months). Male to female ratio was

5:3 (Table 1). Seven cases made a complete recovery. In spring 2014, the eighth case (case H) continued to receive outpatient medical care.

On average 18 close contacts were identified per case (range: 7–33). According to the Irish guidelines [9], relevant close contacts received rifampicin chemoprophylaxis. Pregnant contacts received prophylaxis with intramuscular (IM) ceftriaxone. Among the total number of close contacts identified for all cases, 36 family members were identified as close contacts to more than one case. These family members therefore received chemoprophylaxis repeatedly during the outbreak period. Delay between rounds of chemoprophylaxis in these close contacts varied. For some (close contacts in case A and C), the gap was one year. For others (close contacts in case B and H), the gap was three years.

Laboratory results

All eight cases were diagnosed by detection of *N. meningitidis* serogroup B DNA in extracts of clinical specimens by PCR. Two of the eight cases yielded a *N. meningitidis* isolate from a normally sterile site. Case B had *N. meningitidis* diplococci identified and cultured from CSF on lumbar puncture and case H had *N. meningitidis* isolated from blood culture. Both isolates were identified as B:4:NT/P1.4/NT with the *porA* genotype 7–2,4 (B:4;P1.7–2,4), and were sequence type (ST)-6697 of the ST-41/44 clonal complex by MLST (ST-6697(cc41/44)). Both were sensitive to cefotaxime, ciprofloxacin, penicillin, rifampicin, and sulfadiazine.

TABLE 2

Summary of control measures for an outbreak of invasive meningococcal disease in an extended Irish Traveller family, Ireland, 2010–2013

At risk population identified (N=123)	Clinic X	Clinic Y	Clinic Z	Total
Number invited to attend clinics	31	61	31	123
Number that attended	32 ^a	53	27	112
Number that received chemoprophylaxis	32	53	27	112
Ciprofloxacin	32	52	27	111
Rifampicin	0	1 ^b	0	1
Total number eligible for 4CMenB vaccine	29	43	21	93
Total number that received 4CMenB vaccine	29	36 ^c	20 ^d	86
Number that reported side effects	7	0	0	7
Facial swelling, nausea, vomiting	1	0	0	1
Sore arm at injection site	6	0	0	6

4CMenB: four component meningococcal B vaccine.

^a An additional child, who lives permanently with relatives in this region, attended this clinic as opposed to another clinic site with the rest of their immediate family. Consequently, this child was given chemoprophylaxis and vaccination.

^b Ciprofloxacin contra-indicated.

^c Six family members declined to attend for vaccination; one other family member did not attend and was not followed up as they were almost 24 years-old.

^d One child was unwell at the clinic and did not receive the vaccine.

Characterisation of *N. meningitidis* DNA extracted from the remaining six non-culture diagnosed cases determined two of them to be also ST-6697 (cc41/44) with the *porA* genotype 7–2,4 (cases C and E). Analysis of clinical extracts from two additional non-culture confirmed cases (cases A and F) did not yield a complete MLST profile, so it was not possible to officially assign a clonal complex to these. However, based on their partial MLST profiles (case A: 5 alleles, case F: 4 alleles) and consultation with www.pubmlst.org/neisseria, they are very likely to be ST-41/44 complex meningococci and we have putatively assigned them as so. Also, it was not possible to assign a clonal complex to the isolates from the remaining two cases (cases D and G) due to insufficient MLST data.

Antimicrobial susceptibility results from the EMBU confirmed that the predominant outbreak strain was sensitive to cefotaxime, ciprofloxacin, penicillin, rifampicin, and sulfadiazine.

Control measures for the outbreak

Following the notification of each case, control measures were promptly carried out as per national guidance. These included dissemination of information and administration of appropriate chemoprophylaxis to close contacts. As the outbreak was first recognised in one HSE area (HSE East), information was initially disseminated to local GPs and the Traveller Health Network facilitated by Pavee Point, the Traveller education and development group. Further investigations were initiated to determine whether all cases were caused by the same strain and whether specific risk factors could be identified to account for this cluster. A local outbreak control team (OCT) reviewed the cases.

When additional cases in other HSE areas were identified a national OCT was convened.

The national OCT reviewed all cases linked to the outbreak, ascertained the epidemiological links and risk factors for infection, assessed control measures already implemented and considered what further measures were required. Representatives from Departments of Public Health covering the affected HSE areas, the HPSC, the EMBU and the National Immunisation Office (NIO) participated. Expert opinion was also obtained from a consultant in paediatric infectious diseases.

In June 2013, after the initial national OCT meeting, the following actions were undertaken: (i) an alert was sent to all departments of public health informing them of the outbreak and advising increased awareness of possible further cases; (ii) further characterisation of the outbreak strain/s of *N. meningitidis* was carried out by the EMBU; (iii) communication with the manufacturers of a new four-component protein-based meningococcal serogroup B (4CMenB) vaccine was established to determine availability of this vaccine in Ireland and (iv) immunological testing of cases was requested to determine if there was any underlying susceptibility to infectious diseases. Tests to identify immunological susceptibility to IMD were subsequently carried out on four of the cases. No immunological deficit was detected.

In November 2013, after the eighth case of IMD (case H) was notified it was agreed to offer all members of the extended Irish Traveller family, regardless of whether they had been a contact of an actual case or not, additional simultaneous chemoprophylaxis with ciprofloxacin. Ciprofloxacin is recommended as first line chemoprophylaxis for close contacts of cases of

IMD in the United Kingdom (UK) [19]. As this medication is a single dose treatment compliance can be directly observed. Following recommendations from the independent national expert group (the National Immunisation Advisory Committee (NIAC)), it was also agreed that all members of the extended family aged two months to 23 years inclusive and relevant close contacts of case H would be offered the 4CMenB vaccine, in accordance with manufacturer's immunisation schedule [20].

On 18 December 2013, three clinics were organised to implement these actions. In addition a posterior pharyngeal swab was taken from all family members who attended. In total, 123 family members were invited to attend, of which 112 (91%) attended (Table 2). Of the 11 who did not attend, eight did not attend on the day but subsequently received rifampicin. Three other family members did not attend and did not receive chemoprophylaxis. Of those who did attend, one family member attended an alternative clinic to the one this person was invited to. In December, vaccine was administered at only one clinic site. Children less than two years of age received antipyretic medication on site, in accordance with NIAC recommendations, due to the reported increased risk of post-vaccination fever in this age group. Due to logistical difficulties, the administration of vaccine to eligible family members who attended the remaining two clinics was deferred until January 2014. In total 86 family members, 92% of the 93 who were eligible, received their first dose 4CMenB vaccine by end January 2014.

Arrangements were made with local GPs to complete the vaccination course for the extended family members who required further vaccinations. Public health doctors provided vaccination for the remaining relevant extended family members. Two cases and a number of accompanying family members had relocated to England. Consequently, colleagues at Public Health England agreed to follow up those households.

One family member reported nausea, vomiting and facial swelling after receiving ciprofloxacin and 4CMenB vaccine simultaneously. However, these symptoms resolved spontaneously without additional treatment. When followed up by telephone interview, six family members reported sore arms at the injection site on the day after vaccination. All reported symptoms resolved within three days of onset.

Carriage study results

Posterior pharyngeal swabs were obtained from 112 family members of whom 62 were male (Table 3).

On PCR 15 (13%) swabs were positive for *N. meningitidis* and 14 of these were also culture positive. Thirteen (12%) swabs were PCR positive for serogroup B. Eight of the serogroup B positive isolates were identified as B:4:NT/p1.4/NT:ST6697(cc41/44), seven were found to have por A genotype 7–2.4 on whole genome

sequencing. Five of the 15 positive swabs were from people aged less than 24 years, nine were from those aged between 25 and 39; one positive swab was from a person whose age was unknown. Ten family members with a positive swab result for *N. meningitidis* lived in a trailer/caravan and ten lived in households of eight people or more. All eight family members with B:4:NT/p1.4/NT:ST6697(cc41/44) isolates had previously received chemoprophylaxis. Antimicrobial sensitivity testing indicated that the eight B:4:NT/p1.4/NT:ST6697(cc41/44) isolates were sensitive to cefotaxime, ciprofloxacin, penicillin, rifampicin, and sulfadiazine.

Discussion

This report describes an outbreak of *N. meningitidis* serogroup B in an extended Irish Traveller family over a three and a half year period. While outbreaks of IMD are more commonly associated with serogroup C disease [5,21], prolonged community based and institutional outbreaks of serogroup B disease have been described [22,23]. Control measures in these outbreaks differed. In a prolonged serogroup B outbreak in northern France (2003–2005), an unlicensed monovalent outer membrane vesicle vaccine from Norway was thought to be effective and was administered initially to high risk groups and subsequently to all the population [22]. In contrast, in a prolonged university outbreak in the United States (2008–2010), health promotion and ciprofloxacin chemoprophylaxis were the control measures used. Vaccination was not used as a control measure because a serogroup B vaccine was not licenced in the United States at that time [23]. Family clusters have also been described in the literature but their time duration is considerably shorter than our outbreak [24].

Transmission of *N. meningitidis* occurs through droplet spread of respiratory secretions or saliva [25]. On average 10 to 15% of the population are colonised with *N. meningitidis* at any given time and the duration of carriage can vary from days to months [26,27]. When cases continue to occur, despite appropriate management of contacts, possible reasons for ongoing transmission of *N. meningitidis* include; immunological susceptibility, poor compliance with chemoprophylaxis, resistance to chemoprophylaxis, unidentified close contacts and overcrowding. We considered each of these as a possible cause of this outbreak continuing.

The association between immunological factors, host genetics and IMD is well recognised [28,29]. However, in this outbreak the four cases who underwent comprehensive immunological tests had normal immunological results; therefore immunological susceptibility was not thought a likely cause of increased incidence or ongoing transmission.

Compliance of contacts with the recommended four dose schedule of rifampicin chemoprophylaxis could not be independently verified. Consequently, in order

to ensure compliance, directly observed treatment with ciprofloxacin was provided at the clinics. Antimicrobial susceptibility results from the EMBU confirmed that the predominant outbreak strain was sensitive to cefotaxime, ciprofloxacin, penicillin, rifampicin, and sulfadiazine. These results are in line with previous findings that IMD-associated strains of *N. meningitidis* which exhibit decreased susceptibility to cefotaxime, ciprofloxacin, penicillin, rifampicin and sulfadiazine are currently very rare in Ireland (data not shown).

There were on average 18 close contacts identified per case. This number is high when compared with an average of 7.6 contacts per case reported from a 2012 series of 38 IMD events in four regions in Ireland (data not shown). As this is a larger number of contacts than usually identified, it remains possible that not all close contacts were identified. Despite multiple interviews with the family to ensure identification of as many true close contacts as possible, it was recognised that re-colonisation from a wider network remained a possibility.

A carriage rate of *N. meningitidis* of 13% was identified. This rate may have been affected by the large number of family members who had received chemoprophylaxis previously ($n=68$), especially those who received it in the preceding month, $n=18$. In the general population highest carriage rates of *N. meningitidis* are seen in the 15 to 24 years-old age group [27]. The 2011 Irish census identified 22.4 years as the average age of an Irish Traveller compared with 36.1 years for the general Irish population [30]. In this outbreak associated cohort, 83 (74%) of the participants in the carriage study were less than 25 years of age and 22 (20%) were aged between 15 and 24. It is conceivable that cases in this outbreak had many opportunities to mix with young adult relatives. However, results from the carriage study reveal that only three (14%) of the 22 participants aged between 15 and 24 years demonstrated *N. meningitidis* carriage, compared with nine (45%) of the 20 participants aged between 25 and 40 years. The higher rate of carriage in the older age group is higher than expected when compared with previously undertaken carriage studies which revealed carriage rates of 8.8% in individuals aged 25 and older (data not shown).

Traditionally Irish Traveller family size is larger than that of the general population of Ireland. In 2011, 26.9% of Traveller women reported to have five or more children compared with 2.6% of the general population [30]. Irish Travellers often live in houses or caravans in crowded conditions [30]. Household crowding and attendance at crowded venues such as clubs and parties are recognised as risk factors for carriage of *N. meningitidis* and IMD [31,32]. Irish Travellers also frequently attend large family gatherings where there are large groups of people in close proximity for a prolonged period of time. This is supported by the large number of close contacts identified in the outbreak cases. From the carriage study two thirds ($n=10$, 67%) of the identified *N. meningitidis* carriers lived in a

TABLE 3

Characteristics of individuals who were swabbed for a carriage study, outbreak of invasive meningococcal disease among an extended Irish Traveller family, Ireland, 2010–2013 ($n=112$)

Characteristics of family members swabbed	Swabs ($N=112$) n (%)
Sex	
Male	62 (55)
Female	49 (44)
Not specified	1 (1)
Age groups in years	
0–9	44 (39)
10–19	35 (31)
20–29	13 (12)
≥30	19 (17)
Not specified	1 (1)
Household size^a	
2–7 residents	48 (43)
≥8 residents	56 (50)
Accommodation type	
House	67 (60)
Trailer/caravan	45 (40)
Not specified	0 (0)
Self-reported previous contact of meningococcal case	
No	32 (29)
Yes	80 (71)
If yes, previous chemoprophylaxis?	
Yes	68 (85)
No	12 (15)

^a Only numbers of persons with available information are shown.

trailer or caravan. Census information documents from 2011 identify that 12.3% of Irish Travellers live in trailers or caravans overall [30]. Therefore a larger proportion than expected of *N. meningitidis* carriers lived in a trailer or caravan which is likely to consist of one or two rooms in total. As the carriage study also identified that two thirds ($n=10$, 67%) of positive carriers live in households of eight or more, in contrast to 8.32% of the total Irish Traveller population [30], an element of household crowding is likely to be present.

Additional previously reported factors associated with carriage of *N. meningitidis* include personal and passive cigarette smoking and recent or concomitant respiratory tract infections or influenza [32–34]. Cigarette smoking rates are higher among Irish Travellers than in the general population (52.5% vs 29%) [6,35] and therefore it is possible that cigarette smoking could be associated with this outbreak. However, as information on cigarette smoking is not currently collected on enhanced surveillance for IMD the rate of cigarette smoking for the outbreak family could not be confirmed. Neither information on cigarette smoking nor exposure to cigarette smoking was requested during the carriage study undertaken in December 2013.

Previously it has been difficult to control serogroup B IMD due to the absence of a vaccine. This has changed recently with the approval in 2014 by the Food and Drug Administration of a vaccine for the United States (Trumenba from Pfizer) [36]. Earlier, in January 2013, a four component Men B vaccine, referred to here as 4CMenB (Bexsero from Novartis), was licensed by the European Medicines Agency for use against serogroup B disease [37]. 4CMenB vaccine contains three antigens, factor H binding protein, neisserial adhesion A and *Neisseria* heparin binding agent. In addition, the vaccine contains PorA (serosubtype P1.4) from the outer membrane vesicle of the New Zealand strain 98/254. These antigens affect meningococcal survival, function and virulence [38]. As the outbreak strain was identified as B:4:P1.7–2,4:ST-6697 (cc41/44), we were confident that the use of the vaccine would be beneficial to this outbreak.

The Meningococcal Antigen Typing System (MATS) is an enzyme-linked immunosorbent assay (ELISA) designed to evaluate expression of 4CMenB target proteins and to evaluate their cross reactive potential. Strains are predicted to be covered by 4CMenB if they carry the porA p1.4 epitope or meet a minimum threshold of reactivity to *Neisseria* heparin-binding antigen (NHBA), factor H binding protein (fHbp) or *Neisseria* adhesin A (nadA) in the MATS ELISA, known as the positive bactericidal threshold (PBT) [39]. The case B isolate (which was identified as the outbreak strain) has undergone MATS testing and results predict this strain is covered by any of porA (p1.4), NHBA or fHbp peptides it carries.

The decision by two of the affected HSE regions to delay administration of the vaccine until January 2014 was not thought to have impacted on control measures. The affected family members received ciprofloxacin chemoprophylaxis and therefore short-term eradication of *N. meningitidis* was likely. The duration of eradication of *N. meningitidis* after receipt of chemoprophylaxis can vary from weeks to months [40]. Subsequently, the administration of 4CMenB in January was thought likely to provide longer-term immunity.

To our knowledge, this outbreak was the first time 4CMenB vaccine was used in an outbreak situation in Europe. In December 2013, this vaccine was used in an outbreak of serogroup B disease at Princeton University in the United States when in excess of 5,000 students and staff received this vaccine [41]. More recently in March 2014, the University of California, Santa Barbara vaccinated ca 9,000 students and staff in response to four confirmed cases of serogroup B disease in the university [41].

Conclusion

This is a unique outbreak of IMD caused by *N. meningitidis* serogroup B in an extended Irish Traveller family across three regions of Ireland over a period of three and a half years. This outbreak continued despite

instigating all appropriate control measures on multiple occasions. From descriptive epidemiological data and the carriage study, the most likely risk factor identified for this ongoing outbreak was overcrowding. We hope that the combined use of directly observed ciprofloxacin chemoprophylaxis, in addition to vaccination with 4CMenB will be successful in halting this outbreak. We continue to monitor this family for new cases and strive to ensure that all relevant family members complete the recommended vaccine schedule. To date there have been no further cases of IMD in this family.

This outbreak highlights the importance of recording ethnicity as part of the enhanced surveillance information collected on all cases of meningococcal disease. We recommend the gathering of ethnicity data for key notifiable diseases including IMD. We also recommend that consideration be given to the use of 4CMenB vaccine for vulnerable groups living in similar crowded conditions.

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Conflict of interest

None declared.

Authors' contributions

All authors contributed to the gathering and analysis of the information. Lois O'Connor and Mary Ward wrote the first draft of the manuscript in consultation with the outbreak control team. All authors read and critically revised the first as well as subsequent drafts to this manuscript and approved the final version.

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Genomic analyses of *Francisella tularensis* strains confirm disease transmission from drinking water sources, Turkey, 2008, 2009 and 2012

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Waterborne epidemics of tularaemia caused by *Francisella tularensis* are increasingly reported in Turkey. We have used whole genome sequencing to investigate if *F. tularensis* isolated from patients could be traced back to drinking water sources. Tonsil swabs from 33 patients diagnosed with oropharyngeal tularaemia in three outbreaks and 140 water specimens were analysed. *F. tularensis* subsp. *holarctica* was confirmed by microagglutination and PCR in 12 patients and five water specimens. Genomic analysis of three pairs of patient and water isolates from outbreaks in Sivas, Çorum, and Kocaeli showed the isolates to belong to two new clusters of the *F. tularensis* B.12 genetic clade. The clusters were defined by 19 and 15 single nucleotide polymorphisms (SNPs) in a multiple alignment based on 507 *F. tularensis* genomes. One synonymous SNP was chosen as a new canonical SNP (canSNP) for each cluster for future use in diagnostic assays. No SNP was identified between the genomes from the patient–water pair of isolates from Kocaeli, one SNP between the pair of isolates from Sivas, whereas the pair from Çorum differed at seven SNPs. These results illustrate the power of whole genome sequencing for tracing *F. tularensis* patient isolates back to their environmental source.

Introduction

Tularaemia is a bacterial zoonosis caused by *Francisella tularensis*, a small Gram-negative coccobacillus. The dose required for human infection depends on the transmission route; less than 10 bacteria may be sufficient for infection via inhalation or the subcutaneous route, while infection via the oral route requires greater than 10⁶ bacteria [1,2]. Several clinical forms of tularaemia (including ulceroglandular/glandular, oculoglandular, oropharyngeal and respiratory) may develop, depending on the infection route. Although ulceroglandular

tularaemia is most frequently reported in the literature, the clinical form most frequently seen in Turkey is oropharyngeal tularaemia [3,4]. Epidemiological surveys have revealed a seasonal pattern of the disease in Turkey, where it is endemic in large areas. Most reported cases occur during the winter (December to March), frequently resulting from ingestion of infected food or contaminated water [3,5].

Two main subspecies of *F. tularensis* are clinically important: the most virulent (subsp. *tularensis*) is found only in North America, while the less virulent (subsp. *holarctica*) occurs in most of Eurasia and North America [6,7]. *F. tularensis* subsp. *holarctica* can be further divided into four major genetic clades (B.4, B.6, B.12 and B.16) based on clade-specific canonical single nucleotide polymorphisms (canSNP) markers [8,9]. The distribution of these genetic clades in the waterborne outbreaks in Turkey seems to be widespread [10].

Four documented outbreaks of tularaemia occurred in Turkey between 1936 and 1953; then, following an epidemiologically silent period, it re-emerged in 1988 and its incidence has strongly increased in the past 20 years [11,12]. From 2005, when tularaemia became a notifiable disease in Turkey, to 2012, several outbreaks and in total 4,773 human infections were reported [13,14]. During that time, the three outbreaks studied here of oropharyngeal tularaemia occurred in the Kocaeli (n=35), Çorum (n=54) and Sivas (n=89) areas. Epidemiological investigations indicated that drinking water was the source of infection, but the genotypes and precise sources remained uncertain. In this study, we investigated possible genetic links between *F. tularensis* subsp. *holarctica* strains isolated from patients and drinking water in the same areas and at the same time.

Methods

Tularaemia diagnostics, isolation of bacteria and DNA preparation from clinical and water samples

The outbreaks in the Çorum, Sivas and Kocaeli provinces (Figure 1) commenced on 21 January 2008 (duration approximately two month), 22 February 2009 (duration approximately three month) and 24 March 2012 (duration approximately 40 days), respectively. All tonsil swab specimens collected from index patients were subjected to culturing, RT-PCR and serology. The samples were cultured for 4 to 10 days on cysteine heart agar supplemented with VCNT inhibitor (6 mg/L vancomycin, 15 mg/L colistin, 10 mg/L trimethoprim and 25 mg/L nystatin, Becton Dickinson, Sparks, US). Later during the outbreaks, laboratory verification of tularaemia was based on microagglutination (*F. tularensis* antigen, Becton Dickinson, Sparks, United States (US)) using patient sera [15]. In total, 33 tonsil swab samples and 221 serum samples from patients were analysed. All samples and cultures were handled in a Class III safety cabinet (Labconco, US).

In addition, early during the outbreaks, 51, 34 and 55 specimens of water (0.3 to 1.5 L) were collected from unchlorinated drinking water sources or village fountains in the Çorum, Sivas and Kocaeli areas, respectively. They were transported at +4 °C to Kocaeli University, filtered through cellulose acetate membranes (pore size, 0.22 µm), and filters were placed on cysteine heart agar supplemented with VCNT inhibitor for culturing. Water filtration was carried out in a Class II safety hood and cultivation was performed in a separate laboratory under Class III biological safety conditions.

All analyses, including serology, culturing and RT-PCR of clinical samples and culturing of filtered water, were performed during the outbreaks and without delay upon arrival of the samples at the laboratory.

PCR amplification, detection and identification of *Francisella tularensis* strains

For identification of colonies from clinical tonsil-swabs, DNA was extracted from the colonies and PCR using primer and probe sets targeting *ISFtu2* was done as previously described [13]. Negative and positive controls (the latter consisting of 10-fold dilutions of *F. tularensis* subsp. *holarctica* LVS; NCTC 10857) were amplified in parallel [13]. Colonies cultivated from the filters were identified as *F. tularensis* by agglutination test using specific antibody (*F. tularensis* Antisera, Becton Dickinson, Sparks, US). A PCR assay targeting RD1 was used to identify subspecies of *F. tularensis* isolates from both the tonsil swab and water samples [16].

Whole genome sequencing

Genome sequencing on DNA extracted from the three pairs of patient and water isolates was performed using

Nextera XT DNA preparation kit (Illumina, San Diego, US) and 150 bp paired-end libraries on an Illumina MiSeq instrument. The reads were mapped against the *F. tularensis* subsp. *tularensis* FSC200 [17] genome using Bowtie2 [18] to evaluate the fraction of *Francisella* in the samples. Samples containing a mixture of other bacteria were filtered using MIRA v4.0 mirabait [19]. The filtered reads were subsequently assembled de novo using ABySS [20], and the overall genome coverage was 130-fold (95 contigs). GenBank accession number for the sequences are: FDC200:JPPLOooooooooo, FDC201:JPMTooooooooo, FDC202:JPPMooooooooo, FDC203:JPMUooooooooo, FDC204:JPSVooooooooo and FDC205:JPSWooooooooo.

In silico screening of *Francisella* strains

Genome sequences were initially screened *in silico* by CanSNPer software [21] using published canSNP markers. The markers B.4, B.6, B.10, B.11, B.12, B.13 [22], B.20, B.21 [9], B.26 [23] and B.33 [24] were needed to describe the genotypes in a schematic (canonical SNP) phylogenetic tree. Multiple genome alignment with 507 archived *F. tularensis* strains were used to identify unique SNPs and develop new specific canSNPs for the two clusters detected (B.67 and B.68, see Results), as previously described [25]. The archival genomes represent the available genetic and geographical diversity within *F. tularensis* subspecies *holarctica* from Asia, North America, and Europe.

Genome alignment and phylogenetic tree

A multiple genome alignment of the six *Francisella* genomes obtained in the outbreaks and the 74 most closely related archived *Francisella* genomes was generated by concatenation of a number of pairwise alignments where each strain was aligned against the reference strain FSC200 [17] using progressive MAUVE [26]. The neighbour-joining phylogenetic tree with complete deletion was constructed in MEGA6 [27].

Results

Epidemiology

A total of 178 patients were diagnosed with laboratory-verified tularaemia in the three outbreaks (Figure 1). Pharyngotonsillitis and cervical lymphadenopathy were the most frequent clinical findings in these patients. Epidemiological investigations indicated that drinking water was the likely source of infection. Accordingly, *F. tularensis* subsp. *holarctica* was successfully isolated from cultures of throat swabs collected from 12 patients among 33 index cases investigated. In the 140 drinking water samples, growth of *F. tularensis* subsp. *holarctica* colonies was identified in five samples. One sample pair (one patient and one water isolate) for each of the three outbreaks were selected for phylogenetic analysis.

FIGURE 1

Location of the focal waterborne tularaemia outbreaks in the provinces of Kocaeli, Çorum and Sivas, Turkey, 2008, 2009, and 2012 (n =178 cases)



The number of human tularaemia cases (n) and date of initial case are indicated.

Whole genome sequencing

Six strains were sequenced in total, obtained from one sample pair (one patient and one *F. tularensis*-contaminated water sample) for each of the three outbreaks. Whole genome multiple alignment and CanSNPer analysis showed that all of them grouped in the B.12 clade [8] (Figure 2A). In comparative analyses with whole genome sequences of *F. tularensis* strains of global origin, an isolate from the tonsil of the patient in Kocaeli (FDC204) and the corresponding water isolate from the contaminated water source (FDC205) formed a separate new cluster with identical genome sequences (Figure 2B). The strain most closely related to this cluster was a Swedish strain isolated in 2003 (FSC374), differing at 20 SNPs from the FDC204–5 group (Figure 2B). Other close relatives have been isolated 1967 in Slovakia and 2009 in Hungary (Figure 2B). The other two patient–water pairs, from Sivas and Çorum, were assigned to another new cluster separated by 26 SNPs from its most closely related strain (FSC930, isolated in 1961 in Bulgaria). The water isolates from Sivas and Çorum were identical. However, the patient isolate from Sivas (FDC201) differed from the corresponding water isolate (FDC200) at one SNP at the whole genome level, while the patient isolate from Çorum (FDC203) differed at seven SNPs from the Çorum water isolate (FDC202).

Identification of unique canonical SNPs (canSNPs)

Nineteen and 15 SNPs, respectively, uniquely identified the FDC200–3 and FDC204–5 clusters in the multiple alignment with 507 archived *F. tularensis* genomes.

One synonymous SNP was chosen as a new canSNP for each cluster (B.67 and B.68, respectively). The genomic positions of the B.67 and B.68 SNPs, relative to the *F. tularensis* subsp. *tularensis* SCHU S4 genome (GenBank ID AJ749949.2) [28], were 975,050 and 857,235, respectively. The ancestral base for both B.67 and B.68 was C, and the derived base was A and T, respectively. It was not possible to resolve the two patient–water pairs in the FDC 200–3 cluster, representing strains associated with the outbreaks in Çorum and Sivas, because the sequenced water isolates from these areas (FDC202 and FDC200, respectively) were identical at all aligned positions.

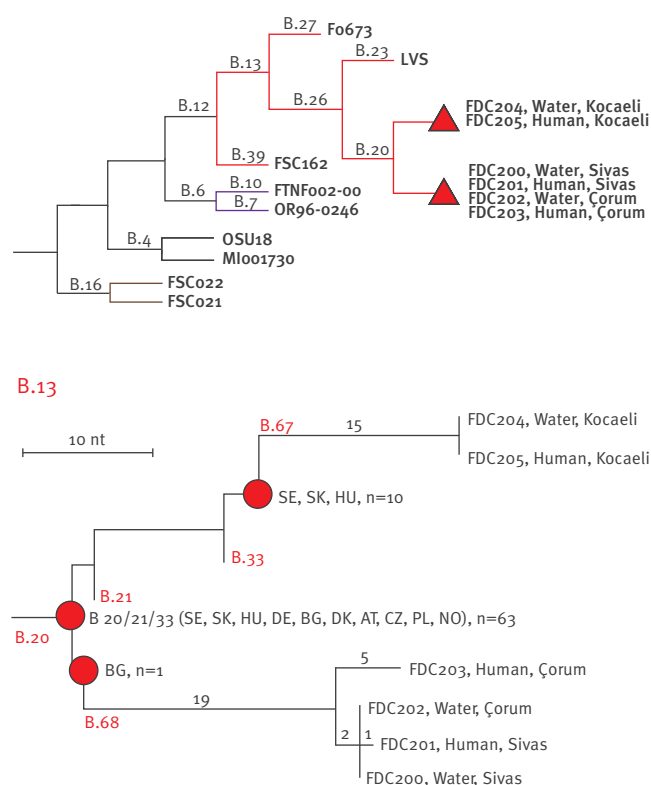
Discussion

Oropharyngeal tularaemia is a significant waterborne disease in Turkey, where more cases were recorded in 2011 than in all European Union countries combined [5]. Better epidemiological tools can help identify the most important transmission chains resulting in human infections in the region and may ultimately aid in preventing the disease.

Because *F. tularensis* exhibits very little genetic variability, high-resolution analytical methods such as whole genome sequencing are appropriate for genetic typing in epidemiological investigations of tularaemia outbreaks and for source-tracing [29,30]. Initial canSNP analysis of the six included isolates revealed that they all belonged to the *F. tularensis* genetic clade B.12, which dominates in Europe between Scandinavia and the Black Sea [7,8,23–25]. The higher resolution

FIGURE 2

Phylogenetic trees showing genetic relationships of the sequenced *Francisella tularensis* genomes, 2008, 2009 and 2012 (n=6)



A. Schematic *F. tularensis* subsp. *holarctica* phylogenetic tree. Names at the branch tips represent sequenced reference strains for each clade, respectively [24]. The red triangles indicate the sequenced *Francisella* genomes from the outbreaks.

B. SNP phylogeny based on the six outbreak *Francisella* genomes and the 74 most closely related archived genomes (red circles). The new CanSNPs (developed in this study) B.68 and B.67, defining the two clusters FDC200–3 and FDC204–5, respectively, and numbers of unique SNPs defining the clusters are indicated. Outbreak strains are indicated by name and origin.

AT: Austria, BG: Bulgaria, CZ: Czech Republic, DE: Germany, DK: Denmark, HU: Hungary, NO: Norway, PL: Poland, SE: Sweden, SNP: single nucleotide polymorphism, SK: Slovakia.

provided by whole genome sequencing showed that the analysed isolates formed two novel genetic clusters separated by at least 20 and 26 SNPs from all archived *F. tularensis* genome sequences. Their distinctions were also confirmed by in silico screening of a large reference collection of *F. tularensis* genomes using the canSNPs (B.67 and B.68) developed in this work to define the two new genetic clusters. The Kocaeli cluster contained two strains with identical genome sequences (isolated from a patient and a water sample), strongly suggesting a common source. The other cluster contained the paired patient–water isolates from Çorum and Sivas. Surprisingly, the genome sequences of the two water isolates from Çorum and Sivas were identical, implying that caution is needed when using genetic data to infer direct epidemiological links. In addition, the human isolate from Çorum represented a subgroup differing at seven SNPs from the water isolates from Çorum and Sivas. Assuming that the source of the human infection

was the water sampled in Çorum, the water must have been contaminated by multiple genetically distinct *F. tularensis* possibly from different sources, e.g. several dead rodents.

Isolating *F. tularensis* from environmental specimens by cultivation is very difficult, mainly because it grows slowly and fastidiously, hence overgrowth of environmental background flora is a major problem. In this study the bacterium was isolated by filtering samples of cold spring water collected during the winter through cellulose acetate membranes before cultivation on selective media. Very low background growth was recorded, probably because the sampled water was pure spring water, which is oligotrophic and thus supports low background bacterial populations.

The isolates analysed in this study were obtained from three locations, 280 to 798 km apart, in central Anatolia and north-western Turkey (Figure 1). No tularaemia outbreaks were reported in these locations before 2004 [1,8]. Ingestion of contaminated water appears to have been the main cause of the focal outbreaks, as in previously reported Turkish outbreaks [13,14]. The patients were using water from the same reservoir (which was unchlorinated at the time of the outbreaks) for both drinking and other needs. The water inlet supplying the used reservoirs in these areas was not piped and the canals supplying drinking water were not covered, leaving the water open for contamination from environmental sources. Thus, it is reasonable to assume that upstream water became contaminated by small rodents killed by tularaemia [5].

The results presented here suggest that although the required infectious dose of *F. tularensis* in humans by the oral route may be high, contaminated drinking water poses a substantial risk to human health. Thus, the often cited bioterrorism potential of *F. tularensis* may not be solely restricted to aerosol distribution, and intentional contamination of drinking water may be an underestimated risk.

In conclusion, whole genome sequencing of outbreak strains confirmed an epidemiological link between drinking water and three outbreaks of human tularaemia in Turkey. The genomic epidemiology approach is particularly powerful for genetically monomorphic bacterial pathogens such as *F. tularensis*.

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Conflict of interest

None declared.

Authors' contributions

All of the authors collaborated in the presented work. AK, MF, AS, MT, HŞ defined the research theme. AK, MF, AS AL, CÖ, KM, LÖ analysed data, interpreted results and wrote the draft manuscript. AJ gave conceptual advice and participated in manuscript writing. All authors have contributed to, seen and approved the manuscript.

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WHO member states adopt global action plan on antimicrobial resistance

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At the 68th World Health Assembly on 25 May, delegates from World Health Organization (WHO) member states endorsed a global action plan [1] to tackle antimicrobial resistance. The goal of the plan is to ensure successful treatment and prevention of infectious disease by securing fair, affordable access to effective antimicrobial agents to those who need it most.

The plan sets out five objectives:

- to improve awareness and understanding of antimicrobial resistance;
- to strengthen surveillance and research;
- to reduce the incidence of infection;
- to optimise the use of antimicrobial medicines;
- to ensure sustainable investment in countering antimicrobial resistance.

The plan mentions that ‘In the European Union alone, a subset of drug-resistant bacteria is responsible annually for some 25 000 deaths, with extra health-care costs and lost productivity due to antimicrobial resistance amounting to at least €1500 million [2].’ The plan provides a framework for national action plans to combat antimicrobial resistance. Through adoption of the plan, WHO member states all agreed to mobilise resources to ensure the plan’s implementation and commit to have national action plans in place by 2017.

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