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HUMAN H5N1 INFECTIONS:  
SO MANY CASES — WHY SO LITTLE KNOWLEDGE?

Angus Nicoll  
European Centre for Disease Prevention and Control, Stockholm, Sweden

This month edition contains an account of clusters of H5N1 infection in humans in Azerbaijan [1]. The account is doubly rare: It describes the first occasion where the source is seemingly wild birds. Reading what happened is reassuring as the people infected had probably killed and defeathered infected swans. I.e. this was not casual exposure to wild birds but rather qualitatively similar to when humans are intimately exposed to sick domestic poultry, which remains the most potent risk factor (one recent analytic study came up with an odds ratio of 29 [2]).

The account is also rare as a peer-reviewed investigation of a cluster of human H5N1 infections. Since reporting began in 2004 there have been 218 confirmed cases in ten countries, mostly in small clusters and WHO has published some details of nearly every one [3,4]. However the number of underpinning analytic investigative reports are embarrassingly small: Consequently little more is known now than in 1997 about an infection that seemingly remains hard for humans to acquire, but is highly lethal when they do (48 of the 74 cases in 2006 died) [3-5]. The only multi-country review has very little information on how transmissions take place and what are the risks, apart from getting too close to sick domestic poultry [6]. For example we still do not really know the reality or rate of asymptomatic and mild human cases around these clusters. While it is stated that there is no evidence that such cases have occurred, a more accurate statement would be that there are hardly any relevant serological data, but what little exists is consistent with few such cases, though equally there are epidemiological data that suggest the opposite [5,6,7]. Equally we are probably underestimating the extent of person to person transmission, which does not matter too much since what must be spotted is whether transmission is becoming more efficient, i.e. when clusters are enlarging in size or duration. Seemingly they are not – yet [4,6].

None of this should be seen as a criticism of any individuals, national health authorities or any single organisation. It is a collective failure but one that must be overcome. Investigations of emerging zoonoses are difficult anywhere. They require simultaneous and coordinated investigations of human and animal cases by joint teams, plus environmental sampling which is difficult even in well-resourced countries [8]. Poor affected communities can be reluctant to be open with officials and investigators as they fear punishment or adverse economic consequences (culling without adequate compensation) [1]. Usually there are multiple confounding exposures which need careful analysis (was the infection from a chicken, poultry products, the environment or another human?). Considerable stamina may be needed as sometimes there are good plans for investigation but they are not implemented after the drama of the outbreak passes. Serological testing of those exposed is incorrectly regarded as a possible research procedure to be done later rather than an important and urgent investigation, consequently it is almost never completed. The academic process does not always help. It can encourage investigators to hold on to ‘bird flu’ does not encourage governments to allow immediate openness.

Usually the problem is not that countries are reluctant to forward information, but rather that the required field investigations are not being done to generate the data in the first place. Having a practical guide to investigations would help and WHO and its Regions are now developing one while ECDC is doing the same for the European Union. Universal use of these and forwarding the results would then allow WHO to populate a global database, at least for newly identified clusters.

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If existing public health measures and anti-virals are to be most effective, countries will need to have fast answers to some important questions from field investigations.
data rather than forward them to WHO and the rare anecdote will be published while the tedious reality will not. Reports that HSN1 could be acquired from eating uncooked duck blood or bathing in canals in Viet Nam are memorable [6,9]. But there have been no analytic studies of these cases taking into account the frequency of these exposures in the population [5]. Unfortunately most of the countries where the first cases have occurred do not have traditions of analytic field investigation and the high profile of ‘bird flu’ does not encourage governments to allow immediate openness. Usually the problem is not that countries are reluctant to forward information, but rather that the required field investigations are not being done to generate the data in the first place. Having a practical guide to investigations would help and WHO and its Regions are now developing one while ECDC is doing the same for the European Union. Universal use of these and forwarding the results would then allow WHO to populate a global dataset, at least for newly identified clusters.

Is the above complaint important or simply one public health person wanting things to be done properly? It is important. This month, the World Health Assembly (16-25 May, Geneva) agreed that implementation of the new International Health Regulations be brought forward. This step was driven by the pandemic threat and the need for early detection and prompt and competent investigation of the first pandemic cases. This is not just to isolate the pandemic strain but also so that WHO’s Rapid Response and Containment tactic could be deployed to stamp out or reduce transmission. Modelling suggests there would only be a short window for newly identified clusters.

four years ago, de Valk and colleagues determined both the need for and feasibility of a European network on Listeria infections in humans [1]. The network was envisioned as a way to strengthen surveillance in individual countries by harmonising microbiological methods and providing epidemiologic tools for investigations. The results of their survey were clear: respondents felt that such a network would aid in the detection and investigation of outbreaks, and that it could be based on existing national surveillance systems [1]. There has been considerable institutional support for developing a European Listeria network, and in response to planning efforts, Listeria surveillance has improved in several countries [2]. However, the network has yet to be realised.

This issue contains a series of articles that, along with earlier reports from the Netherlands and England and Wales, highlights the current status of Listeria surveillance in Europe, documents trends in the occurrence of the disease, and illustrates the growing need for a European surveillance network. In 2002, reported incidence of listeriosis in Europe ranged from 0 to 7.5 cases per million inhabitants [1]. The highest rates were reported from countries that had statutory notification of listeriosis and increased yield in reported cases associated with foodborne illness. The relationship between public health investment in surveillance and increased yield in reported cases was subsequently demonstrated in the Netherlands. Although listeriosis is not a notifiable disease in the Netherlands, implementation of more active surveillance in January 2005 has resulted in a 43% increase in the reported incidence of listeriosis [2].

Three papers in this issue report a full spectrum of trends from national surveillance data. In France, the incidence of listeriosis declined from 4.5 cases per million inhabitants in 1999-2000, to 3.5 cases per million inhabitants during the period from 2001-2003 [3]. In Finland, the number of reported cases varied markedly by year from 1995-2004, but there was no clear trend and the mean annual incidence was 7.5 cases per million inhabitants [4]. In Germany, incidence increased from 2.6 cases per million inhabitants in 2001 to 6.2 cases per million inhabitants in 2005,
with most of the increase occurring among people over 60 years of age [5]. A similar increase in listeriosis among people over 60 years of age occurred in England and Wales from 2001-2004 [6].

In France and Finland, routine serotyping and molecular subtyping by pulsed-field gel electrophoresis (PFGE) resulted in the detection of several case clusters and common-source foodborne outbreaks. However, few isolates in Germany were serotyped or subtyped by PFGE, and no foodborne outbreaks were identified. The importance of routine molecular typing of Listeria isolates for outbreak detection and investigation was further highlighted by the two outbreaks reported in this issue from Switzerland, where the incidence of listeriosis has been stable but relatively high, and the United Kingdom, where incidence has been increasing [7,8]. Although both were identified because of a regional clustering of cases, rapid characterisation of an outbreak strain facilitated both investigations. Ultimately, isolation of the outbreak strains from implicated food items confirmed the source of the contamination [8].

The national experiences with listeriosis surveillance summarised in this issue suggest that across much of Europe, rates of listeriosis may be increasing or remaining stable at relatively high levels. In Germany, the increasing proportion of highly susceptible persons in the population was cited as a contributing factor to the increased incidence of listeriosis [5]. Indeed, across Europe the population is ageing and the prevalence of cancer increased by 40% between 1990 to 2002 [10]. However, a growing at-risk population should not inevitably increase the public health burden of listeriosis. Where rates of listeriosis are declining, such as in France, this appears to be the result of extensive surveillance efforts to define the scope of the problem, followed by active collaboration between public health officials, food regulatory officials and food producers to reduce the levels of contamination in the food supply [3].

European food safety standards will help establish consistent approaches to the control of Listeria in ready-to-eat foods. However, implementation of these standards will still require extensive collaborations at the national level. Reliable surveillance data on listeriosis are a foundation upon which effective collaborations are built. Strengthening surveillance in individual countries by harmonising microbiological methods and providing epidemiologic tools for investigations will be a key step in reducing the public health burden of listeriosis, even as the population at risk grows. Thus, the need for a European surveillance network for Listeria has never been greater.

References


Editorial

Infection Risks from Water in Natural and Man-Made Environments

Gordon Nichols
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People can catch diarrhoeal diseases from contamination of both natural and man-made environments with human or animal faeces. Young children are more likely to be susceptible to the agents and to be exposed. While some diarrhoeal diseases acquired in childhood can be relatively mild and give some protection as an adult, others are more severe. The two papers presented in this issue of Eurosurveillance describe, on the face of it, unremarkable small outbreaks; one, from Chikwe Ihekweazu et al, linked to exposure to a stream contaminated with Escherichia coli from animal faeces [1]; the other, from Melanie Jones et al, to exposure to a water feature contaminated with Cryptosporidium parvum from either animal or human faeces [2].

Rivers, lakes and streams are known to harbour enteric and other pathogens derived from sewers, animal waste, the environment or through contamination by the bathers themselves. Outbreaks associated with recreational activity in these environments have been reported in developed countries [3-6]. However, the burden of illness associated with these sources as most disease is assumed to be acquired in a sporadic fashion. The source of contamination can sometimes be determined by tracking the specific pathogen type causing the illness to an upstream host. In the absence of such evidence, source tracking methods using indicator organisms or other markers [7-9] remain unreliable. While risk assessment can be used to reduce exposure to contamination in some situations it cannot prevent all disease. Although what we would like to have is good prevention of most disease, in practice assessing risks from recreational waters is both complicated and beset by local difficulties. The World Health Organisation has produced guidelines [10] that provide appropriate approaches to controlling infectious diseases and other risks. Epidemiological studies have used bathing trials to examine the relationship between microbial indicators of water quality and diaries of symptoms kept by the participating volunteers [11]. Bathers at a number of sites were exposed to swimming in the sea or not and then followed up symptomatically, and the symptoms compared to microbiological measurements of faecal contamination of the water. Such studies have shown that there is a relationship between exposure to faecal pollution in general and faecal enterococci in particular and the burden of reported gastrointestinal symptoms. Retrospective cohort studies have also been used to examine the risks from recreational bathing with similar results [12].

Such studies suffer from a variety of methodological criticisms. There is scepticism about the relationship between reported diarrhoeal symptoms and the acute diarrhoeal diseases that are diagnosed by laboratory detection of causative agents. Most human gastrointestinal pathogens exhibit a seasonal distribution. The
human and animal faecal inputs are likely to exhibit a different distribution. Because of this the relationships between pathogen and indicator when measured throughout the year are likely to vary by orders of magnitude. As an example Norovirus is the commonest cause of human gastrointestinal disease but is not thought to derive from animals. There will therefore be some relationship between human faecal contamination and norovirus infection whereas there will not be with animal contamination. As with disease burden studies related to drinking water [13] this approach has to generalise from the conditions within the local environment of the study to a general assessment. Despite this, there is a need to set new standards and the levels established from bathing studies have been revised for this purpose.

A new EU Directive [14], was published by the European Union on 4 March 2006 and entered into force 20 days later on 24 March. Under the Directive the tests for bathing waters are simplified to E. coli and intestinal enterococci, instead of 19 different tests used previously. It will classify beaches as either ‘excellent’, ‘good’, ‘sufficient’ or ‘poor’. The extra classification of ‘sufficient’ quality comes below ‘excellent’ and ‘good’ but still allows a beach to qualify as a bathing water and the standards have been raised so that the estimated health risk to bathers is reduced. There will be more tests carried out more frequently when a beach is classified as ‘poor’ or only ‘sufficient’. Information on water quality will be provided on all bathing beaches to show the quality of recent tests. Under this new regime it is hoped that infections linked to recreational activity will be reduced. MEPs voted on 18 January 2006 to allow the new standards to replace the existing 1976 Directive. This bathing water management programme was introduced over a 13 year period, starting in 2008.

There is a difference between recreational water activity in natural and man-made environments. In recent years there has been an increase in outbreaks of infectious diseases associated with public water features of various types [15-21]. It seems that there are factors in the design of many of these features that increase the risks of people, particularly children, being infected. Outbreaks in other countries have involved Shigella sonnei [20], norovirus [19], legionnaires disease [17] and Pontiac fever [18]. The microbiology of such water features and the treatment of the water within them have received little attention. There have been a number of recent outbreaks linked to recreational water features in England and Wales caused by cryptosporidium. There was also a large outbreak of cryptosporidiosis at the Seneca State Park sprayground (an interactive water feature) in New York State, USA, in August 2005 which affected an estimated 3000 people. Cryptosporidium was found in two water storage tanks that supplied water to a water spray attraction.

A variety of private and municipal water features are being developed that allow people, particularly young children, to play in them. These may present risks to the populations using them if they are not designed and operated correctly. These features differ from swimming pools in potentially having a greater burden and variety of environmental contamination and requiring a high water turnover that puts a burden on any treatment processes.

Interactive water features are usually located outdoors and include fountains, shallow pools, vertical pressure jets, overhead sprays and showers. Children can run around in and easily drink the water. The area is usually designed to collect the water from the feature and return it to an underground holding tank. The water jets are operated by pumps that utilize their water from a holding tank. The features are often fitted with control valves that enable operation to be varied either manually or via an automatic programme. The holding tank should be sized to ensure that there is adequate water available to operate the feature and there should be a separate system for water treatment. These features pose a high risk of microbiological contamination and transmission of infection to children. The filtration systems need to be well designed and managed to remove cryptosporidium oocysts that can escape from the environment and from fountains’ shoes and bodies. Additionally, the disinfection should be sufficient to inactivate bacterial and viral pathogens. The microbiological quality of water at the feature’s spouts of the feature should be to the same standard as swimming pool water and should be checked at least monthly (BS PAS 39:2003). Water should ideally be mains water that is not re-circulated. In all cases the UK Water Supply (Water Fittings) Regulations 1999 apply. Where re-circulation is required treatment should involve filtration and disinfection as occurs with swimming pools. With interactive water features the risk of cryptosporidium infection may be the same as, or greater than that from swimming pools. The use of UV treatment to reduce the risk of cryptosporidiosis is recommended. There should be clear signs indicating that the water is not for drinking, and alternative sources of safe drinking water should be readily available.

Interactive water features may suffer from environmental contamination, including domestic and wild animals and birds, and people can occasional cause accidental fouling with vomit or faeces. In these instances the contaminated water should be diverted to drain and the pool cleaned. These features need to automatically make-up water lost by evaporation and filter backwashing. Some plant rooms may be located underground and these should be well designed for housing all equipment and ensuring the safe delivery and storage of chemicals. Water from these features should not be used to top up other pools as this could lead to contamination and an outbreak [22].

There are a variety of municipal water features including decorative pools and fountains, that have not been designed for bathing but which are used for this purpose in hot weather, often by children. These pools can involve the same problems as interactive water features and may also have inadequate filtration and disinfection. Such pools should be designed to make it difficult for children to use them as recreational play areas. Indoor features such as fountains have also been responsible for outbreaks of legionellosis, which probably reflects higher water temperatures. Outbreaks in other countries have involved Shigella sonnei [20], norovirus [19], legionnaires disease [17] and Pontiac fever [18]. The microbiology of such water features and the treatment of the water within them have received little attention. There have been a number of recent outbreaks linked to recreational water features in England and Wales caused by cryptosporidium. There was also a large outbreak of cryptosporidiosis at the Seneca State Park sprayground (an interactive water feature) in New York State, USA, in August 2005 which affected an estimated 3000 people. Cryptosporidium was found in two water storage tanks that supplied water to a water spray attraction.

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Increasing evidence of outbreaks linked to both recreational waters and decorative water features

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Increasing evidence of outbreaks linked to both recreational waters and decorative water features
and management. All such features should be formally assessed for microbiological risks, including legionella, during the design stage and ensure that treatment is adequate for minimising the risks to the public. Risk assessment should involve a public health microbiologist. The risk assessments should be reviewed at regular intervals and at least every two years. The principal microbiological risks are cryptosporidiosis resulting from inadequate filtration, legionellosis resulting from inadequate disinfection, and bacterial and viral infections also resulting from inadequate disinfection. In addition to infection risks there needs to be assessments of other risks such as slipping, drowning [36] and disembowelment [37,38]. Disinfection and filtration systems must be well maintained and monitored. Measures should be in place to minimise faecal contamination, especially from footwear, and to minimise potential for children to drink of the water. Recent outbreaks indicate that there is a risk of litigation if water features are found to be the cause of an outbreak. If an outbreak is associated with such a feature, consideration should be given to pool closure and drainage until the pool can be shown to be safe.

What should we conclude from these two papers about the risks of infection? There is increasing evidence of outbreaks linked to both recreational waters and decorative water features. While the source of contamination on bathing beaches may be contaminated of the sea from rivers, the diffuse sources from small streams can be important in contributing to contamination and may be missed in an investigation. As for interactive water features, the design and use must be carefully managed to ensure that outbreaks resulting from children drinking water contaminated with cryptosporidium are avoided.

References

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Mandatory notification of listeriosis began in France in 1999. Enhanced public health surveillance, including routine molecular characterisation of Listeria monocytogenes strains, epidemiologic follow-up of cases, and collection of food samples, has improved the sensitivity of outbreak detection and response. The incidence of listeriosis declined from 4.5 cases/million in 1999-2000 to approximately 3.5 cases/million during the period 2001-2003. Clinical, demographic and microbiological characteristics of listeriosis in France remained stable during this time period. Maternal-fetal infections accounted for 24% of all cases. Serovar 4b accounted for 49% of cases and 60% of case clusters. The incidence of listeriosis in France has declined and is now lower than in several other European countries.

Methods

**Definitions**

A case is defined by isolation of L. monocytogenes from a patient with a clinically compatible illness. A case is considered maternal/neonatal (MN) when it involves a pregnant woman, a miscarriage, a stillbirth, or a newborn under one month old. When L. monocytogenes is isolated from both the pregnant women and her newborn child, it is counted as a single case. If a case fits none of these groups, it is considered non-maternal/neonatal (non-MN). Patients are considered to be at risk if they have an underlying pathology weakening their immune system, including cancer, blood malignancy, organ transplant, chronic haemodialysis, liver failure, diabetes, HIV, treatment with cytolytic or corticosteroid immunosuppressants.

**Collection of data**

Information collected on the notification sheet includes the department of residence of the patient, his/her age, the clinical form of disease, the possible existence of an underlying pathology at the time of listeriosis diagnosis and whether the patient was still living at the time of the follow-up. The food questionnaire was administered face to face or by telephone, and includes a list of food items, thought to be likely sources of Listeria, that may have been consumed in the two months before onset of illness. This includes food items previously identified as vehicles in outbreaks, and foods which have been previously found to be contaminated by L. monocytogenes, and are consumed uncooked. Given the wide variety of such products existing in France, this list is not exhaustive and does not take into account certain products rarely consumed in France. The questionnaire consists of a list of 76 items in four categories: seafood products (seven items): fish-based products, smoked fish, shrimp and shellfish; vegetable products (three items): packaged lettuce, packaged prepared raw vegetables and soy sprouts; dairy products (34 items): 33 cheeses and unpasteurised milk; cold cuts, cooked meats and meat-based products (32 items): pâté, ham, sausages, meat products with gelatine, poultry-based products.

**Analysis of strains by the CNR**

Strains are confirmed to be L. monocytogenes; in parallel they are characterised by serotyping and analysis of DNA macrorestriction profiles according to standard protocols [1,3,4]. Isolates with indistinguishable ApaI and Ascl macrorestriction profiles, based on visual comparison of banding patterns, were considered to be the same pulsovar. Sensitivity to antibiotics is studied using diffusion techniques. Resistance is confirmed using MIC determination (E-test method) by the antibiotic resistance national reference centre
Management of listeriosis clusters

A cluster is defined as the occurrence of at least three listeriosis cases over a period of 14 weeks and involving strains of the same pulsovar. After a cluster is detected by the CNR, the ‘listeria group’ is informed. It is composed of representatives of InVS, the CNR for Listeria, and the Ministries of Agriculture, Health and Economy (consumers protection directorate). The InVS analyses patient data (notification forms and food questionnaires) and, if cases appear to be linked, the ‘listeria group’ develops and coordinates the investigations needed to identify a possible common source and implement appropriate measures to prevent the spread of disease.

Quality of the monitoring system

The sensitivity of the system for reporting diagnosed cases of listeriosis has been estimated at 87% by the capture-recapture method comparing data collected in 2001 by mandatory notification with the data reported by the EPIBAC network [5].

Results

Epidemiological characteristics

The annual incidence of listeriosis in France decreased in 2001 [TABLE 1] and stabilised at the lower rate with 3.4 cases/1 000 000 inhabitants notified in 2003. From 2001 to 2003, the mean number of cases per year was 206, with 49 maternal/neonatal cases and 157 non-maternal/neonatal cases. Between 2001 and 2003, the mean regional incidence was 3.0 cases/1 000 000 inhabitants (range 0–5.1) [FIGURE 1]. Regional distribution of cases did not differ significantly from one region to another during the study period. A seasonal effect, with an increase of cases during summer, was observed [FIGURE 2]. The mean number of cases for the period from May to August was 262, compared with 118 cases for the other four month periods.

Maternal/neonatal forms

Maternal/neonatal forms represented 24% of cases. From 2001 to 2003, 48 cases of neonatal or fetal mortality were notified: 21 miscarriages, 19 stillbirths and 8 newborns who died within the first 48 hours after birth.

Non-maternal/neonatal (non-MN) forms

The non-MN forms were more common in males (57% of cases from 2001 to 2003). This was observed both for subjects with no risk factors, and for subjects with underlying listeriosis risk factors. The presence or absence of underlying pathology was known for 462 of the 471 non-MN cases notified between 2001 and 2003. Of these 462 patients, 337 (73%) had an underlying risk factor, 56 were affected by pathology not considered to be a risk factor for listeriosis, and 69 had no known pathology at the time of admission to hospital. One hundred and seven patients died within 10 days after diagnosis (20% lethality).

Table 1

Principal characteristics of listeriosis cases identified by disease notification and isolate submission to the National Listeria Reference Laboratory, France, 1999–2003

<table>
<thead>
<tr>
<th>Risk factor</th>
<th>1999</th>
<th>2000</th>
<th>2001</th>
<th>2002</th>
<th>2003</th>
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<tbody>
<tr>
<td>No detectable pathology</td>
<td>32</td>
<td>32</td>
<td>16</td>
<td>27</td>
<td>26</td>
</tr>
<tr>
<td>Known risk</td>
<td>140</td>
<td>130</td>
<td>107</td>
<td>113</td>
<td>117</td>
</tr>
<tr>
<td>Other pathology</td>
<td>20</td>
<td>32</td>
<td>18</td>
<td>22</td>
<td>16</td>
</tr>
<tr>
<td>Unknown</td>
<td>10</td>
<td>5</td>
<td>3</td>
<td>3</td>
<td>3</td>
</tr>
</tbody>
</table>

Sex

<table>
<thead>
<tr>
<th>Sex</th>
<th>1999</th>
<th>2000</th>
<th>2001</th>
<th>2002</th>
<th>2003</th>
</tr>
</thead>
<tbody>
<tr>
<td>Female</td>
<td>78</td>
<td>74</td>
<td>55</td>
<td>70</td>
<td>73</td>
</tr>
<tr>
<td>Male</td>
<td>124</td>
<td>125</td>
<td>89</td>
<td>95</td>
<td>89</td>
</tr>
</tbody>
</table>

Serovar (all forms)

<table>
<thead>
<tr>
<th>Serovar (all forms)</th>
<th>1999</th>
<th>2000</th>
<th>2001</th>
<th>2002</th>
<th>2003</th>
</tr>
</thead>
<tbody>
<tr>
<td>% serovar 4b</td>
<td>51</td>
<td>54</td>
<td>42</td>
<td>56</td>
<td>47</td>
</tr>
<tr>
<td>% serovar 1/2a</td>
<td>24</td>
<td>30</td>
<td>33</td>
<td>22</td>
<td>27</td>
</tr>
<tr>
<td>% serovar 1/2b</td>
<td>29</td>
<td>13</td>
<td>22</td>
<td>17</td>
<td>22</td>
</tr>
<tr>
<td>% serovar 1/2c</td>
<td>5</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>5</td>
</tr>
</tbody>
</table>

Source: InVS Notifiable Disease Reports

Figure 1

Listeriosis incidence per million inhabitants, by region, France, 2001–2003

Source: InVS Notifiable Disease Reports

Figure 2

Number of listeriosis cases by month, France, 2001–2003

Source: InVS Notifiable Disease Reports

Figure 2

Number of listeriosis cases by month, France, 2001–2003

Source: InVS Notifiable Disease Reports

Table 1

Principal characteristics of listeriosis cases identified by disease notification and isolate submission to the National Listeria Reference Laboratory, France, 1999–2003

<table>
<thead>
<tr>
<th>Region</th>
<th>1999</th>
<th>2000</th>
<th>2001</th>
<th>2002</th>
<th>2003</th>
</tr>
</thead>
<tbody>
<tr>
<td>Case notified</td>
<td>269</td>
<td>263</td>
<td>188</td>
<td>220</td>
<td>209</td>
</tr>
<tr>
<td>Case with strain submitted to CNR</td>
<td>254</td>
<td>250</td>
<td>186</td>
<td>218</td>
<td>199</td>
</tr>
<tr>
<td>Incidence per 1 000 000 inhabitants</td>
<td>4.5</td>
<td>4.4</td>
<td>3.1</td>
<td>3.6</td>
<td>3.4</td>
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<tr>
<td>Clinical form</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-maternal-neonatal</td>
<td>202</td>
<td>199</td>
<td>144</td>
<td>165</td>
<td>162</td>
</tr>
<tr>
<td>Maternal-neonatal</td>
<td>67</td>
<td>64</td>
<td>44</td>
<td>55</td>
<td>47</td>
</tr>
<tr>
<td>Deaths</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adult</td>
<td>47</td>
<td>34</td>
<td>46</td>
<td>35</td>
<td>26</td>
</tr>
<tr>
<td>Neonatal and fetal</td>
<td>19</td>
<td>23</td>
<td>13</td>
<td>22</td>
<td>13</td>
</tr>
<tr>
<td>Total</td>
<td>66</td>
<td>57</td>
<td>59</td>
<td>57</td>
<td>39</td>
</tr>
</tbody>
</table>

Source: InVS Notifiable Disease Reports
Strain analysis

From 2001 to 2003, the CNR received 603 L. monocytogenes strains which accounted for 98% of cases notified to the INVS [TABLE 1]. Approximately half of all strains were serovar 4b (4b: 49% of the strains, 1/2a: 27%, 1/2b: 20%, 1/2c: 4%, 3a and 3b <1%). Distribution of serovars by clinical form [TABLE 2] showed that:

i) a higher proportion of strains of serovar 1/2b were isolated from MN infections (27%) than from non-MN forms (18%) (p=0.02),

ii) a higher proportion of strains of serovar 1/2c were isolated from non-MN forms (5%) than from MN forms (1%) (p=0.03),

iii) among non-MN forms, serovar 4b was frequently isolated from CNS infections. Within the strains of serovar 4b, 42% were isolated from CNS infections. Values for the other serovars were 32.8% (1/2a), 28.1% (1/2b), and 23.8% (1/2c), respectively (p = 0.0006).

In 2003, 92 pulsovars were identified with between one and 15 strains per pulsovar, resulting in a total discrimination index (that is, the probability that two randomly chosen strains would have different pulsovars) of 0.980. Three resistance profiles were observed. Three strains were resistant to ciprofloxacin (serovar 1/2a), two were resistant to tetracycline and trimethoprim (serovar 1/2a) and one was resistant to tetracycline alone (serovar 1/2b).

<table>
<thead>
<tr>
<th>TABLE 2</th>
</tr>
</thead>
</table>

**Distribution of 603 strains of Listeria monocytogenes from human cases, by serovar and clinical form, France, 2001-2003**

<table>
<thead>
<tr>
<th>Serovar</th>
<th>1/2a n (%)</th>
<th>1/2b n (%)</th>
<th>1/2c n (%)</th>
<th>4b n (%)</th>
<th>Other n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-maternal-neonatal [n=462]</td>
<td>134 (29)</td>
<td>82 (18)</td>
<td>21 (5)</td>
<td>221 (48)</td>
<td>4 (1)</td>
</tr>
<tr>
<td>Central nervous system infection [n=165]</td>
<td>44</td>
<td>23</td>
<td>5</td>
<td>93</td>
<td>0</td>
</tr>
<tr>
<td>Bacteraemia [n=270]</td>
<td>83</td>
<td>56</td>
<td>15</td>
<td>112</td>
<td>4</td>
</tr>
<tr>
<td>Focal infection [n=27]</td>
<td>7</td>
<td>3</td>
<td>1</td>
<td>16</td>
<td>0</td>
</tr>
<tr>
<td>Maternal-neonatal. [n=141]</td>
<td>29 (21)</td>
<td>38 (27)</td>
<td>1 (1)</td>
<td>73 (52)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Total. [n=603]</td>
<td>163 (27)</td>
<td>120 (20)</td>
<td>22 (4)</td>
<td>294 (49)</td>
<td>4 (&lt;1)</td>
</tr>
</tbody>
</table>

Source: National Listeria Reference Laboratory

Investigation of clusters

Between 2001 and 2003, the CNR reported 25 clusters. The median size of clusters was five cases, with a range of 3-14 cases per cluster. Fifteen clusters (60%) were due to serovar 4b strains. Cases involved in clusters represented 26% of notified cases during this period. Analysis of the cases’ food consumption histories identified suspected food vehicles for several clusters and triggered further investigations and control measures at production site. In three outbreaks, a L. monocytogenes strain of the case-associated pulsovar was identified in a food product consumed by several cases, confirming the food item as the source of the outbreak.

Discussion

The incidence of listeriosis in France decreased substantially from 1987 through 1997 after control measures were implemented by the food industry in response to several large outbreaks [5]. Following implementation of mandatory notification, incidence has further declined from 4.5 cases/1 000 000 in 1999-2000 to approximately 3.5 cases/1 000 000 during the period 2001-2003. As this reduction concerns all forms of listeriosis (targeted or not by food recommendations) this further decline is likely due to a reduction of exposure to contaminated product [5]. This reduction could be the consequence of more effective recall of contaminated products from the marketplace following a directive issued by the Ministry of Agriculture in 1998 that standardised recall procedures. In addition, mandatory notification with routine, standardised collection of food consumption histories has allowed prompt cluster investigations and identification of the source of the outbreaks, thus reducing the number of cases exposed to implicated products.

The proportion of maternal-neonatal cases which had declined significantly from 1987 (51% of all cases) to 1997 (24% of all cases) has not changed since 1997. Other clinical and demographic characteristics of listeriosis in France (male to female sex ratio, case fatality rate) have also remained stable. Serovar 4b remains the most common serovar and accounts for a disproportionate share of case clusters and central nervous system infections, as previously noted [6].

Conclusion

Thanks to the joint efforts of food producers, government and health authorities, the incidence of listeriosis in France, which in the 1980s exceeded that of other industrialised countries, is now lower than in several other European countries and is at the same level as the United States, which applies a ‘zero-tolerance policy’ for Listeria in foods [7,8]. Although there have been fewer episodes than in preceding years, there remains a risk for case clusters and outbreaks due to contaminated food [9].

Acknowledgments

We would like to thank Craig Hedberg, DMI, INVS for his valuable contribution to improving the manuscript’s English.

References

We analysed the surveillance data from listeriosis cases notified to the Finnish National Infectious Diseases Register between 1995 and 2004 and describe our recent experience in investigating clusters of listeriosis cases. The number of annual cases varied between 18 and 53 but no trends in incidence were identified (average annual incidence was 7 cases per million inhabitants). Only a few cases affected pregnant women or newborns. Most of the patients were elderly people with non-malignant underlying illnesses; 25% of them died from their infections. By routine sero- and genotyping of the listeria isolates, we detected several clusters; the vehicle for infection was only identified for two outbreaks. At least one quarter of listeriosis cases (78/315) was caused by a certain sero-genotype or closely related genotypes, which have also been found from vacuum-packed cold-smoked or cold-salted fish products. During 2000-2003, Finnish consumers were repeatedly informed about food precautions for risk groups. The information was also given to attending physicians and prenatal clinics.

Methods
Since 1995, physicians in Finland have been obliged to notify culture confirmed cases of listeriosis to the NIDR, which is maintained at the National Public Health Institute (KTL)’s Department of Infectious Disease Epidemiology, and the microbiology laboratories that isolate L. monocytogenes from blood, cerebrospinal fluid, genital tract, newborn, deep puncture, and surgical specimens. Strains of L. monocytogenes must also be sent to KTL’s Enteric Bacteria Laboratory for serotyping and pulsed field gel electrophoresis (PFGE).

L. monocytogenes isolates were serotyped for their O and H antigens by slide and tube agglutination methods, respectively, using commercially available antisera (Denka Seiken Co., Ltd, Tokyo, Japan) according to the manufacturers’ instructions with minor modifications [7]. In situ DNA isolation and macrorestriction analyses by PFGE using the restriction enzyme Ascl were performed as described [7].

When a cluster of listeriosis cases was detected, clinical information (underlying conditions/illnesses and outcome) was collected from the attending physician using a standardised form. In addition, patients or their family members were interviewed by phone about food and drink consumed during the four weeks before the onset of illness. One matched case-control study was performed to identify the potential association between illness and the consumption of a certain food.

Results
Between 1995 and 2004, 18 to 53 cases of listeriosis were identified annually in Finland; 3-10 cases per 1 000 000 inhabitants per year [FIGURE 1, data are based on NIDR notifications]. The average annual incidence rate varied from 2 to 13/1 000 000 inhabitants by region. Of all patients with listeriosis, 57% were 65 years of age or older and 55% were male. Between zero and three cases each year were occurred in pregnant women or newborns.

The most common serotypes were 1/2a (60%) and 4b (23%); only during 1998-1999 serotype 3a was more common than serotype 4b [Table 1, data are based on the 315 strains submitted to the Enteric Bacteria Laboratory]. PFGE types among the strains of serotypes 1/2a and 4b were diverse and no single dominating type was found, whereas PFGE type 71 (‘butter type’); the strain type that was responsible for the high mortality make outbreaks difficult to recognise and investigate, especially for smaller clusters.

To assess the trends in incidence and persons at risk, we analysed surveillance data from listeriosis cases notified to the National Infectious Diseases Register (NIDR) during 1995-2004. We also describe our recent difficulties in investigating clusters of listeriosis cases.
Most of the cases caused by this genotype occurred in 2004 (7/13); with a maximum of two cases per year during 1995-2003. The cases caused by this strain occurred in several regions around Finland.

During 1999-2004, after the outbreak linked to butter, clinical information was collected from 75 cases of listeriosis during three different time periods when infection clusters were suspected [TABLE 2]. Of the positive cultures, 60 (80%) were from blood, five (7%) from cerebrospinal fluid and 10 (13%) from other sources (three from fluid in the abdomen, two from pleural fluid, two from deep puncture, one from pus, one from an abscess and one from urine). Only four cases (5%) occurred in pregnant women or newborns. Almost all patients (67/70) who were not pregnant had at least one underlying illness, but illness was malignant in less than one third of these cases (20/67). A total of 20 patients died; 12 (16%) died within one week after positive listeria culture and 19 (25%) within one month.

Between 7 June 1999 and 15 March 2000, 27 cases of listeriosis were reported, of which 13 were caused by strains of 'fish types'. Of the 27 cases, 25 were included in the case-control study (one newborn and a patient with skin infection were excluded). Three control subjects matched by age, underlying medical conditions and hospital were identified for each case with the help of the attending physicians. Analysis of the 25 cases and 62 matched controls showed no association between illness and consumption of fish products (Odds ratio (OR) 1.7; confidence intervals (CI) 95% 0.6-5.8), and nor did the subanalysis, which included only the 13 cases caused by strains of 'fish types' and their matched controls (OR 1.8; CI 95% 0.4-9.6). However, 17 (68%) of the 25 case-patients and 9 (69%) of the 13 cases caused by strains of 'fish types' had eaten uncooked fish products within the incubation period; most often cold-salted fish. The fish products consumed by the case-patients could not be traced with a maximum of two cases per year during 1995-2003. The cases caused by this strain occurred in several regions around Finland.

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During a short period at the beginning of 2002 (5.1.2002-4.2. 2002) listeriosis was detected in six people, five of whom were from southwest Finland. However, this local cluster of listeriosis cases was caused by strains of two different serotypes and three genotypes (4b-65, 1/2a-96 and 1/2a-253). Five of the six patients were interviewed about food histories but the interviews did not identify any common food.

From 12 November 2003 to 31 December 2004, we attempted to interview all people who had been ill with listeriosis, or if the patient had died, family members of the deceased. We succeeded in interviewing approximately half of the patients (22/42). Genotyping simultaneously revealed two clusters with seven cases each [FIGURE 2: sero-genotypes 1/2a-27 and 4b-56]. The food histories of the people infected by sero-genotype 1/2a-27 were strongly suggestive of cold-salted fish products (four out of five patients cases had consumed these products). During the same period of time, four additional people became ill with listeriosis caused by strains of 'fish types', but they were not interviewed. Only three of the people infected by sero-genotype 4b-56 were interviewed, and no common food history of well known risk foods (raw, unpasteurised) milk and foods made from raw milk, soft cheeses, paté, meat and fish products) was identified.
### Table 2
Clinical and demographic characteristics of 75 listeriosis cases in Finland during three different time periods, 1999-2004

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Source of positive culture</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Blood</td>
<td>22 (81)</td>
<td>6 (100)</td>
<td>32 (76)</td>
</tr>
<tr>
<td>Cerebrospinal fluid</td>
<td>2 (7)</td>
<td>0 (0)</td>
<td>3 (7)</td>
</tr>
<tr>
<td>Other</td>
<td>3 (11)</td>
<td>0 (0)</td>
<td>7 (17)</td>
</tr>
<tr>
<td>Median age, years (range)</td>
<td>69 (0-86)</td>
<td>59 (41-80)</td>
<td>72 (22-91)</td>
</tr>
<tr>
<td>Male sex</td>
<td>14 (52)</td>
<td>4 (67)</td>
<td>22 (53)</td>
</tr>
<tr>
<td>Underlying condition</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Haematologic malignancy</td>
<td>4 (15)</td>
<td>2 (33)</td>
<td>8 (19)</td>
</tr>
<tr>
<td>Solid malignancy</td>
<td>4 (15)</td>
<td>1 (16)</td>
<td>1 (2)</td>
</tr>
<tr>
<td>Solid organ transplantation</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Pregnancy</td>
<td>3 (11)</td>
<td>0 (0)</td>
<td>1 (2)</td>
</tr>
<tr>
<td>Newborn</td>
<td>1 (4)</td>
<td>0 (0)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>No underlying condition (not pregnant)</td>
<td>1 (4)</td>
<td>0 (0)</td>
<td>2 (5)</td>
</tr>
<tr>
<td>Death</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Within 1 week after positive culture</td>
<td>6 (22)</td>
<td>3 (50)</td>
<td>3 (7)</td>
</tr>
<tr>
<td>Within 1 month after positive culture</td>
<td>9 (33)</td>
<td>3 (50)</td>
<td>7 (17)</td>
</tr>
</tbody>
</table>

### Discussion
The annual incidence of listeriosis in Finland has not decreased during the last ten years. Pregnancy related cases are still rare. Most of the persons who became ill with listeriosis were elderly people with underlying illnesses, less than third of which were malignant. A quarter of the case-patients died.

The routine subtyping of listeria isolates by both pheno- and genotypic methods allowed us to identify clusters that might have had a common vehicle and source. Several small clusters were detected. The comparison of typing results of human listeria strains with those obtained from foods may give clues about the implicated food and the interviews may then focus on this type of food. In 2004, three people became ill with listeriosis caused by a sero-genotype that had previously been found in vacuum-packed cold-smoked or cold-salted fish products, or caused by its closely related sero-genotypes [4,7]. In 2002, there were also three such cases, and there were 11 such cases in 2003 and 14 such cases in 1999. Similar linkages between human clusters and fish products without epidemiological association have also been reported from Sweden, Norway and Iceland [8-10]. Based on these human findings, the National Food Agency, the National Veterinary and Food Research Institute, and KTL made several announcements (press release) (three times in 2000 and once in spring 2003) that vacuum-packed cold-salted or cold-smoked fish products may contain *L. monocytogenes*, which may cause listeriosis, especially in people at high risk [11]. In 2000, attending physicians and Finnish prenatal clinics were also given information about food precautions for risk groups [see Box] [12].

In practice, listeriosis cases caused by the same listeria sero- and genotype often occur over a relatively long period of time and are geographically dispersed. To minimise recall bias, food history interviews should be performed as soon as possible after the onset of illness. However, culture findings from human specimens for detailed typing are usually not yet available at that point in time, and without typing results, the cluster cannot easily be recognised. Therefore, if interviews are carried out before typing results are available, it is not possible to include more detailed questions concerning certain foods.

### Box
Current food precautions to reduce the risk of listeriosis in Finland

**General recommendations:**
- Cook all meat thoroughly
- Wash raw vegetables thoroughly before eating
- Keep uncooked meat separate from vegetables and from cooked foods and ready-to-eat foods
- Avoid raw (unpasteurised) milk or foods made from raw milk
- Wash hands, knives, and cutting boards after handling uncooked foods
- Recommendations for persons at high risk:
  - Avoid soft aged cheeses, such as blue cheese, and fresh cheeses
  - Cook left-over food or ready-to-eat food until steaming hot
  - Avoid vacuum-packed cold-salted or cold-smoked fish products

From: [www.ktl.fi](http://www.ktl.fi) and [www.elintarvikevirasto.fi](http://www.elintarvikevirasto.fi)

By performing an analytical epidemiological study, we potentially could show an association between illness and consumption of a certain food item: whether the case-patients are more likely to have consumed certain food in comparison with the controls. In listeriosis outbreaks, the number of cases is usually small, many case-patients die and some are too ill to be interviewed. Matching according to underlying condition may lead to matching by level of exposure, and bias the results to zero (that is, less likely to identify a risk factor) [5,13]. Finding controls with the help of an attending physician can be laborious. Sometimes, the suspected foods are very commonly consumed and it is not possible confirm the association with a relatively small number of study subjects. For the above mentioned reasons, it is often advisable to inform the public, particularly those people at high risk, to avoid certain foods even if there is no evidence of the vehicle or source of infection. Communication between health and food authorities about the typing results of human and food...
isolation of L. monocytogenes from sterile specimens or neonates are reported to the Robert Koch-Institut. Listeriosis—incidence significantly increased from 0.26 per 100,000 inhabitants (217 cases) in 2001 to 0.62 per 100,000 (519 cases) in 2005. The increase only occurred among non-pregnancy-associated cases and was mainly due to a rise in cases in the age group ≥60 years. The highest incidences were observed in neonates and adults ≥70 years. Male cases predominated, except for cases occurring in adults of childbearing age. The overall case fatality rate was 9%. No temporal or spatial clusters of cases were observed and no outbreaks with a common source vehicle were identified. In 46% of the cases malignancies were reported as predisposing factor. Reasons for the increase of listeriosis in Germany remain unclear. The newly implemented surveillance system, and raised diagnostic awareness, cannot explain the particularly high increase in incidence from 2004 to 2005. Increased contamination of common foodstuffs or changes in underlying medical conditions or treatment options may have contributed to the increase. A project for enhanced listeriosis surveillance was begun in 2005 to obtain more detailed information about the clinical course, underlying conditions, medical treatment, knowledge about listeriosis and possible food risk factors from all newly diagnosed cases. For better outbreak detection, a nationwide system for molecular subtyping of listeria strains from humans and food is necessary. Recommendations for prevention should be extended to all risk groups with predisposing conditions.

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Key words: listeriosis, surveillance, increase, epidemiology

Original articles
Surveillance report

Significant increase of listeriosis in Germany - Epidemiological patterns 2001-2005

J Koch, K Stark

Listeriosis has been a mandatorily notifiable disease in Germany since January 2001. Clinical cases with isolation of Listeria monocytogenes from sterile specimens or neonates are reported to the Robert Koch-Institut. Listeriosis—incidence significantly increased from 0.26 per 100,000 inhabitants (217 cases) in 2001 to 0.62 per 100,000 (519 cases) in 2005. The increase only occurred among non-pregnancy-associated cases and was mainly due to a rise in cases in the age group ≥60 years. The highest incidences were observed in neonates and adults ≥70 years. Male cases predominated, except for cases occurring in adults of childbearing age. The overall case fatality rate was 9%. No temporal or spatial clusters of cases were observed and no outbreaks with a common source vehicle were identified. In 46% of the cases malignancies were reported as predisposing factor. Reasons for
Introduction

Listeriosis, caused by *Listeria monocytogenes*, is a foodborne infection of great public health concern due to its clinical severity (resulting in, for example, abortion, septicemia or meningitis) and high case fatality. Most affected by severe disease are people who are elderly or immunocompromised, pregnant women and neonates (younger than four weeks). In recent years, an increase of listeriosis cases including larger outbreaks has been observed in several European countries. In this paper, we report the time trends and epidemiological data of listeriosis cases reported in national surveillance in Germany from 2001 to 2005.

Methods

In Germany, listeriosis has been a notifiable disease since 2001[1, 7]. All cases from whom *L. monocytogenes* is cultured from blood, cerebrospinal fluid, or other usually sterile specimens must be reported to the local public health department by the identifying laboratories. The health departments complete and verify the case information based on the national case definition for listeriosis. Information about clinical signs and outcome is obtained either electronically transmitted to the state health department and from there to the Robert Koch-Institut (RKI), the national public health institute. For quality assurance each individual case report is checked at RKI for plausibility of the laboratory and clinical data according to the case definition. In neonates, the isolation of *L. monocytogenes* from any specimen is notifiable and fulfils the case definition independent of clinical signs and symptoms. According to the case definition data of listeriosis, cases not in neonates are included in the national surveillance database if the infection is laboratory confirmed and clinical disease is present [2]. Until 2001 only cases of congenital listeriosis had to be reported.

Since the beginning of 2004 when the listeriosis case definition was revised, mothers of neonates with listeriosis are also reported (as epidemiologically linked cases), irrespective of their clinical picture or laboratory results. Therefore, the number of pregnancy related listeriosis cases for the years 2001 to 2003 cannot be directly compared with these cases from 2001 to 2003. In addition, the clinical signs and symptoms of premature delivery, flu-like symptoms and fever were added to the list of possible manifestations for pregnancy-associated cases.

Results

Between 1 January 2001 and 31 December 2005, 1519 cases of *L. monocytogenes* were reported to the RKI.

The case numbers significantly increased from 217 cases in 2001 (incidence: 0.26 per 100 000 inhabitants) to 510 cases in 2005 (incidence: 0.62 per 100 000) (p<0.001; z-test). The overall incidence has more than doubled since the introduction of a mandatory notification system of culture confirmed listeriosis cases at the beginning of 2001. From 2001 to 2004 the annual increase of listeriosis cases ranged from 7% to 16%. In 2005, cases increased 72% compared to 2004. No seasonal trends were observed in listeriosis incidence, and no outbreaks were reported. The temporal and spatial distribution of cases, especially during the increase of 2005, did not reveal any clusters suggestive of local outbreaks. Cases could not be linked to any common source or vehicle of infection.

Annual totals for the years 2001 to 2005 demonstrate that the number of pregnancy-associated listeriosis cases (including neonates) showed some fluctuation but no clear trend, while non-pregnancy associated listeriosis (excluding neonates) dramatically increased during this time period [FIGURE 1]. A total of 1294 cases (85%) of all reported 1519 cases were not pregnancy related. Of the non-pregnancy related cases, 76% were in patients aged ≥60 years. Between 2001 and 2005 the number of cases in the age group ≥60 years increased by a factor of 2.6, from 132 to 346 cases, while the case number in the younger group increased by a factor of only 1.7, from 56 to 97 cases. The increase was sharpest in the age group ≥80 years where almost four times as many cases were reported in 2005 (n=86) as in 2001 (n=22). Since 2001, a total of 225 pregnancy-associated cases (including neonates) have been notified, representing 15% of all cases. If we assume that the number of pregnant cases for the years 2001 to 2003 would have been higher if the modified case definition of 2004 had already in place since the beginning of 2001, then we can say that the annual number of pregnancy-associated cases during 2001 to 2005 remained relatively stable.

**FIGURE 1**

Annual number of reported listeriosis cases by patient category and age group, Germany, 2001-2005

Figure 2 shows annual listeriosis incidence by age group and sex for the years 2001 to 2005. The highest incidences are seen in neonates and adults ≥70 years. Neonates show an incidence of 4.2 per 100 000 inhabitants. In neonates, boys (4.9/100 000) were more frequently affected than girls (3.6/100 000). In the age groups 20-29 years and 30-39 years, incidence was higher in women, due to the pregnancy related cases. Of all 126 cases in the age group 20-39 years, 98 (78%) were in women, and 77 (61%) were pregnancy related. Overall incidence increased continuously in the age groups 50-59 years and older and reached 1.2 per 100 000 among those aged ≥70 years (men 1.7/100 000, women 0.82/100 000). In the age groups ≥40 years, the majority of cases are men.

**FIGURE 2**

Age and sex distribution of listeriosis cases, Germany, 2001-2005
According to the case definition, only data from cases with clinical symptoms are presented. To fulfil the definition for a clinically and laboratory confirmed case, it is sufficient if the case shows one of the listed clinical signs, and so data collection about the clinical signs is not comprehensive. In the majority of cases only the leading symptom is reported. In 314 cases multiple responses regarding the clinical signs were given. Among the cases not related to pregnancy (n=1294) the signs and symptoms most frequently reported were meningitis (32%), septicaemia (26%), fever without characteristic organ involvement (31%), abscess (4%), and endocarditis (3%) [TABLE 1].

**Table 1**
Clinical symptoms of non-pregnant listeriosis patients (n=1294), Germany, 2001-2005

<table>
<thead>
<tr>
<th>Clinical symptoms</th>
<th>Number of cases (percentage)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Meningitis</td>
<td>424 (32%)</td>
</tr>
<tr>
<td>Septicaemia</td>
<td>335 (26%)</td>
</tr>
<tr>
<td>Others</td>
<td>235 (18%)</td>
</tr>
<tr>
<td>Localised infection of other organs</td>
<td>120 (9%)</td>
</tr>
<tr>
<td>Abscesses</td>
<td>47 (4%)</td>
</tr>
<tr>
<td>Endocarditis</td>
<td>43 (3%)</td>
</tr>
<tr>
<td>Joint infection</td>
<td>11 (1%)</td>
</tr>
<tr>
<td>Fever</td>
<td>403 (31%)</td>
</tr>
</tbody>
</table>

* Multiple responses possible

Collection of data about pregnant women with listeriosis has improved since the simultaneous notification of these cases was implemented in 2004. From 2001 to 2005 the clinical manifestations of 80 pregnancy associated cases were reported. The proportion of pregnant listeriosis cases for which clinical information was available increased from 68% in 2001 to 2003 to 84% in 2004 and 2005.

The mean annual case number for the period previous to the change of the case definition was about 10, while in 2004 and 2005 about 25 cases annually were reported [TABLE 2]. The most common symptoms and clinical outcomes among pregnant women (n=80) were premature delivery (33%), fever (31%), flu-like symptoms (16%) and miscarriage/abortion (13%).

**Table 2**
Clinical symptoms of pregnant listeriosis patients (n=80), Germany, 2001-2005

<table>
<thead>
<tr>
<th>Clinical symptoms</th>
<th>2001-2003 n=31</th>
<th>2004-2005 n=49</th>
<th>Total n=80</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flu-like symptoms</td>
<td>n (%)</td>
<td>n (%)</td>
<td>n (%)</td>
</tr>
<tr>
<td>Fever</td>
<td>4 (13)</td>
<td>21 (43)</td>
<td>25 (31)</td>
</tr>
<tr>
<td>Septicaemia</td>
<td>1 (3)</td>
<td>1 (2)</td>
<td>2 (3)</td>
</tr>
<tr>
<td>Meningitis</td>
<td>1 (3)</td>
<td>0 (1)</td>
<td>1 (1)</td>
</tr>
<tr>
<td>Miscarriage/abortion</td>
<td>11 (35)</td>
<td>1 (2)</td>
<td>12 (15)</td>
</tr>
<tr>
<td>Premature delivery</td>
<td>2 (6)</td>
<td>24 (49)</td>
<td>26 (33)</td>
</tr>
<tr>
<td>Stillbirth</td>
<td>3 (10)</td>
<td>2 (4)</td>
<td>5 (6)</td>
</tr>
<tr>
<td>Asymptomatic</td>
<td>10 (32)</td>
<td>8 (16)</td>
<td>18 (23)</td>
</tr>
</tbody>
</table>

* Multiple responses possible

In 138 (9%) of all listeriosis cases reported from 2001 to 2005, the patient died. The case fatality was highest in neonates (11%) [TABLE 3]. It was relatively low (0 to 4%) in the age groups between one year and 49 years, but increased to 11% in the age group 50-59 years and 12% in the age group ≥70 years.

**Table 3**
Case fatality rate for listeriosis cases by age group, Germany, 2001-2005

<table>
<thead>
<tr>
<th>Age group (years)</th>
<th>Deaths</th>
<th>Case fatality rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;1</td>
<td>17</td>
<td>11%</td>
</tr>
<tr>
<td>1-19</td>
<td>0</td>
<td>0%</td>
</tr>
<tr>
<td>20-29</td>
<td>1</td>
<td>3%</td>
</tr>
<tr>
<td>30-39</td>
<td>1</td>
<td>1%</td>
</tr>
<tr>
<td>40-49</td>
<td>4</td>
<td>4%</td>
</tr>
<tr>
<td>50-59</td>
<td>16</td>
<td>11%</td>
</tr>
<tr>
<td>60-69</td>
<td>28</td>
<td>7%</td>
</tr>
<tr>
<td>≥70</td>
<td>71</td>
<td>12%</td>
</tr>
<tr>
<td>Total</td>
<td>138</td>
<td>9%</td>
</tr>
</tbody>
</table>

The mean annual listeriosis incidence in the years 2001 to 2005 was 0.37 cases per 100 000 for the whole of Germany. However, substantial geographic variations of the incidence were observed. It ranged from 0.16 cases per 100 000 in the state of Mecklenburg Vorpommern to 0.63 cases per 100 000 in the city state of Bremen. Figure 3 displays the incidence differences by federal state.

Information about the country where the listeriosis had been acquired was available for 1297 cases (85%). In 98% of the cases the infection had most likely been obtained in Germany.

*L. monocytogenes* was detected by culture in 1463 cases (from blood 71%, cerebrospinal fluid 24%, other usually sterile patient...
specimens 4%, material from neonates 2%). Serotyping was only carried out in 5% of cases (n=80). Serotype 1/2a was found in 39 cases, serotype 4b in 38 cases and serotype 1/2b in 3 cases.

Information about the underlying medical conditions of the listeriosis cases cannot be systematically obtained in routine surveillance. In an ongoing project of enhanced listeriosis surveillance we aim to collect such information from all cases. However, information about the underlying conditions or predisposing factors was available for 257 (20%) of the 1294 cases not related to pregnancy. The conditions reported most frequently were malignancies (46%, of which non-haematological malignancies 28%, haematological malignancies 18%), followed by liver cirrhosis (11%), other underlying conditions such as HIV/AIDS, psoriasis, rheumatoid arthritis, collagen vascular disease (11%), immunosuppressive treatment (9%), or diabetes (7%) [TABLE 4].

**Table 4**

<table>
<thead>
<tr>
<th>Underlying disease or condition in non-pregnant patients with listeriosis, Germany, 2001-2005</th>
<th>Number (percentage)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-haematological malignancy</td>
<td>71 (28)</td>
</tr>
<tr>
<td>Haematological malignancy</td>
<td>45 (18)</td>
</tr>
<tr>
<td>Cirrhosis</td>
<td>28 (11)</td>
</tr>
<tr>
<td>Other underlying condition</td>
<td>28 (11)</td>
</tr>
<tr>
<td>Immunosuppressive treatment</td>
<td>22 (8)</td>
</tr>
<tr>
<td>Diabetes mellitus</td>
<td>19 (7)</td>
</tr>
<tr>
<td>Dialysis</td>
<td>12 (5)</td>
</tr>
<tr>
<td>Organ transplant</td>
<td>11 (4)</td>
</tr>
<tr>
<td>Inflammatory bowel disease</td>
<td>10 (4)</td>
</tr>
<tr>
<td>Alcoholism</td>
<td>8 (3)</td>
</tr>
<tr>
<td>Chronic liver disease</td>
<td>3 (1)</td>
</tr>
<tr>
<td>Total</td>
<td>257 (100)</td>
</tr>
</tbody>
</table>

Discussion and conclusions

Listeriosis surveillance data in Germany reveal a continuous increase of cases since 2001, when the national reporting system was introduced. A particularly steep increase was observed in 2005. This trend is mainly due to an increase of non-pregnancy related cases aged ≥60 years, and is most pronounced in the age group ≥80 years. Incidence of non pregnancy related listeriosis is higher among males. A possible explanation is that the number of several predisposing conditions such as malignancies and alcoholic disease in males is likewise higher in males than in females.

It is a common phenomenon that case numbers may increase in the first one or two years after the implementation of a new surveillance system. However, the further rise in listeriosis in Germany in the previous two years cannot be explained by factors such as better acceptance of the surveillance system among laboratories and physicians, or raised diagnostic awareness. In other European countries with a longer history of listeriosis reporting such as England and Wales or the Netherlands similar trends of increasing listeriosis case numbers have been observed [3,4,8]. In conjunction with the data from Germany this indicates a true rise in incidence rather than a surveillance artefact.

Although we cannot rule out the possibility that part of the increase may be caused by enhanced diagnostic awareness of physicians, the data suggest that listeriosis incidence among elderly people has truly increased. The reasons for this, however, remain unclear. It is likely that the proportion of highly susceptible patients (immunosuppressive treatment, medical conditions, etc.) is increasing over time in an aging population [6]. However, this would result in a steady but rather slow increase and cannot explain the significant increase in 2005. It is possible that common foodstuffs were more frequently contaminated with Listeria in recent years. This remains rather speculative since no systematic and representative large-scale food investigations have been performed. However, there is evidence from routine food safety investigations that substantial proportions of different foodstuff may be contaminated by L. monocytogenes (e.g., about 10% of raw meat products in 2005). Unfortunately, serotyping and molecular typing results for L. monocytogenes isolates is only rarely performed in Germany. Therefore, we do not have any laboratory data which would allow to identify (diffuse) listeriosis outbreaks and possibly link isolates from human cases to those from certain foods. Although there is no evidence from the surveillance side that larger outbreaks occurred, the relatively long incubation period makes it difficult to establish epidemiological links between cases and to identify a common food vehicle by epidemiological studies only.

The observation that the number of pregnancy associated cases remained relatively stable while the number of non-pregnancy related cases steadily increased over time might be explained by the fact that risk communication and prevention strategies are already well-established in the risk group of pregnant women. For the other risk groups (high age, immunosuppression, malignancies) intensified education and preventive efforts are required.

The further marked increase of listeriosis in 2005 prompted us to start an enhanced surveillance project for listeriosis. The aim of the project is to obtain detailed and standardised information about the clinical course, underlying conditions, medical treatment, knowledge about listeriosis in risk groups and possible alimentary risk factors from all newly diagnosed listeriosis cases in Germany. In order to gain better insight into the epidemiology of listeriosis, enhanced surveillance and epidemiological studies should be combined with the implementation of molecular typing of isolates from humans and food.

Efforts to educate high risk consumers and thereby reduce their risk of listeriosis should be intensified. The recommendation for the prevention of listeriosis that pregnant women should avoid high risk foods should be continued. Other people with predisposing conditions for listeriosis such as immunocompromised individuals and the elderly should also be informed about possible risk factors and prevention strategies.

References

An outbreak of listeriosis occurred in the Swindon area of the UK in autumn 2003. Five cases were detected in pregnant women. Four of these women were thought to have eaten prepacked sandwiches from a retail outlet in one particular hospital. Sampling at the supplier detected Listeria monocytogenes, which was indistinguishable on molecular testing from the patients’ isolates. Recent changes in UK food legislation should help diminish the risk of further outbreaks/cases such as ours occurring.

Two further cases were then detected in the Swindon area, and so investigations to find a common source continued. A second questionnaire was used, asking in more detail about the types of food eaten within the three months before onset of illness. These revealed that, apart from shopping at major supermarket chains, the only other similarity was that three of the patients had eaten prepacked sandwiches from a single retail outlet within the Great Western Hospital, Swindon which they had attended for antenatal appointments, and a fourth patient had probably eaten them on previous antenatal appointments. This fourth case thought she had eaten them but could not be 100% certain due to the long time period asked in the questionnaire and difficulty remembering.

The EHOs visited the outlet and found sandwiches sold during that period had come from two national suppliers and one local supplier. Daily temperature records for all the refrigerators and between pack of sandwiches measurements had been kept, and the refrigeration records were unremarkable. However, the outlet’s contract with the local supplier had just been terminated and these sandwiches were no longer available for purchase in the hospital.

An outbreak meeting was held and the following actions were taken: active surveillance was initiated by alerting local Consultants in Communicable Disease Control (CsCDC) and microbiology departments, the outbreak was reported in the national communicable disease epidemiological bulletin (CDR Weekly), [13] and the HPA FSML at Colindale was contacted to find out whether any isolates with a similar profile had recently been identified. Case 5 was notified by the local microbiologist and, at the same time, information was supplied by FSML that this was a similar isolate (by typing). Healthcare workers working with pregnant women and neonates in the Swindon area were alerted to the outbreak and the local population was informed via the media (newspaper, radio and television coverage).

The EHOs visited the premises of the local sandwich supplier, and samples of food and environmental swabs were taken for microbiological testing for Listeria. A sample from a brie and cranberry sandwich grew Listeria monocytogenes, as did environmental samples from the premises (chopping boards, sink plug holes and cleaning sponge). On further serotyping and molecular testing, these were shown to be indistinguishable from blood culture isolates from all the patients at the HPA FSML. They were all typed as serotype 1/2, phage type Y, Amplified Fragment Length Polymorphism (AFLP) type III and were indistinguishable by pulsed field gel electrophoresis (PFGE) using AscI, a rare profile in the UK.

This sandwich supplier voluntarily closed down in order to clean the premises thoroughly. The EHOs also visited the supplier that provided meat and cheese for this sandwich maker. Samples were taken but none yielded listeria.

The hospital retail outlet was given advice about the future purchase of sandwiches (see discussion).

Key words: Listeria, Outbreak, Pregnant, Sandwiches
**Discussion**

Listeriosis is not a notifiable disease in the UK, and so it can be difficult to recognise outbreaks early. This outbreak was detected because most of the patients (four out of five) presented to the Great Western Hospital in Swindon or had a link with it. A recent survey of European countries showed that surveillance systems are in operation in 16 of the 17 countries surveyed and that in 10 of these countries the infection is statutorily notifiable [2]. If cases of listeriosis were made notifiable in the UK, all known cases would be reported, which would help to detect outbreaks where cases are scattered throughout the UK.

The incubation period for listeriosis can be long (between 3-70 days) [14] and the food questionnaires used in our outbreak investigation had to cover a period of several weeks. The patients may therefore have had difficulty remembering exactly what they had eaten during this period. A link was, however, established for three of the cases (and possibly a fourth) - these patients all remembered eating prepacked sandwiches bought from a retail outlet in the hospital. No link was found for the fifth case. Two previous outbreak reports have found an association with sandwiches supplied by external contractors within hospitals [15,16]. In Cardiff [15], two cases of listeria septicaemia occurred in immunosuppressed patients who were day cases in the hospital on the same day, and the only food link found was that both had eaten commercially prepared sandwiches supplied by the hospital. These sandwiches were sampled and grew *L. monocytogenes* with serogroup, AFLP type and phage type all indistinguishable from the patients’ isolates. Similarly, four cases of listeriosis occurred in and around the city of Newcastle in a two month period [16]. This outbreak was traced back to a caterer who provided sandwiches for the hospital shop. In our outbreak and the outbreaks in Cardiff and Newcastle [15,16], patients who were at risk (that is, immunocompromised or pregnant) visited the hospital and obtained food that was contaminated with *Listeria*. We consider that providers of food to places with higher than average concentrations of people with lowered immunity, such as hospital retail outlets, providers of food to places with higher than average concentrations of people with lowered immunity, such as hospital retail outlets, should be made aware of the need for the highest possible standards of food hygiene.

In January 2006 new food hygiene legislation came into force in the UK enacting EC Regulations. The new guidelines [17] recommend that food businesses manufacturing ready-to-eat foods, which could pose a risk to public health through the presence or growth of *L. monocytogenes*, should monitor processing areas and equipment for the presence of this organism as part of their sampling plans. In our outbreak and in others [10,11], the environment was shown to be contaminated and may have led to product contamination. The guidelines also recommend that if the food is to be stored before consumption (that is, if it has a shelf life) then *L. monocytogenes* should not exceed 100 cfu/g during this period. If this level cannot be guaranteed, then it should be absent from 25 g when it leaves the food business operator. We hope that these new guidelines will prevent outbreaks such as the one described here.

In conclusion, we report an outbreak of listeriosis that occurred in pregnant women and was associated with the consumption of prepacked sandwiches (ready-to-eat food) from a hospital outlet. However, recent changes in the UK food legislation, if enforced, should diminish the risk and help prevent further cases/outbreaks occurring in similar circumstances.

**Acknowledgements**

Thanks to Jim McLauchlin and Kathy Grant of the HPA FSML for providing typing and information, and to Christina Rattigan (Maternity Unit, GWH) and Linda Wearn (Swindon PCT) for assistance with sending out information to healthcare workers.

**References**


**Table**

**L. monocytogenes, Swindon, United Kingdom, 2003**

<table>
<thead>
<tr>
<th>Case number</th>
<th>Time interval between cases (from DOB of index case)</th>
<th>Symptoms of mother</th>
<th>Sites where <em>L. monocytogenes</em> was recovered</th>
<th>Gestation at time of delivery</th>
<th>Hospital in which baby delivered</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0</td>
<td>Fever</td>
<td>Mother: blood culture Baby: nose and ear swabs</td>
<td>36 wks</td>
<td>Blackpool</td>
</tr>
<tr>
<td>2</td>
<td>+3 days</td>
<td>Fever with flu-like symptoms Previous week had diarrhoea and vomiting</td>
<td>Mother: blood culture</td>
<td>Term</td>
<td>Swindon</td>
</tr>
<tr>
<td>3</td>
<td>+34 days</td>
<td>Unwell, Fever</td>
<td>Mother: blood culture</td>
<td>Twins born at 29 weeks + 2 days</td>
<td>Swindon</td>
</tr>
<tr>
<td>4</td>
<td>+33 days</td>
<td>Fever</td>
<td>Mother: blood culture Baby: blood culture</td>
<td>26 wks</td>
<td>Swindon</td>
</tr>
<tr>
<td>5</td>
<td>-20 days</td>
<td>Nil in mother (Breathing difficulty in baby)</td>
<td>Mother: vaginal swab Baby: blood culture</td>
<td>37.5 wks</td>
<td>Gloucester</td>
</tr>
</tbody>
</table>
Outbreak report

OUTBREAK OF HUMAN LISTERIOSIS ASSOCIATED WITH TOMME CHEESE IN NORTHWEST SWITZERLAND, 2005

J Bille1, DS Blanc1, H Schmid2, K Boubaker2, A Baumgartner2, HH Siegrist3, ML Tritten3, R Lienhard3, D Berner4, R Anderau4, M Treboux5, JM Ducommun5, R Malinverni6, D Genné6, Ph Erard6, U Waespi7

During an eight week period in spring 2005, 10 cases of listeriosis were reported in a small area of northwest Switzerland (150 000 inhabitants). Eight cases were in older immunocompromised patients who became ill with bacteraemia (three deaths), and two cases were in pregnant women who had septic abortion. All cases were due to a serotype 1/2a isolate with one of two pulsotypes found by PFGE. Patient interviews quickly revealed that a locally made and distributed soft cheese (known as ‘tomme’) was the food source responsible for the outbreak. Samples of this cheese, and of butter made in the same factory, revealed Listeria monocytogenes sv 1/2a of the same pulsivar in amounts of 1000-10000 and 10-100 cfu/g, respectively. The prompt suspension of production, the market recall of the product, and a public alert terminated the outbreak. However, two cases of febrile gastroenteritis due to the same strains were reported within 10 days of product recall. The restricted distribution area of the contaminated cheese and the collaboration of local physicians, medical microbiologists and food health services all contributed to a rapid and successful investigation.

This small outbreak of listeriosis reinforces the need for a laboratory-based surveillance system with rapid typing, as well as collaboration between physicians and microbiologists.

Introduction

Human listeriosis is endemic in Europe, with an annual incidence varying between 0.3 and 0.7 cases per 100 000 inhabitants [1]. It has only been 25 years since the recognition that human listeriosis is almost exclusively a foodborne disease, and in this time, many outbreaks of varying extent have been reported, mostly in Europe and North America. The food items most often implicated in outbreaks have been dairy products (milk, soft cheese), meat (pâté, rillettes, sausage and various delicatessen), fish (smoked trout), and vegetables (coleslaw, sweetcorn salad) [2].

Between 1983 and 1987, Switzerland experienced a long-lasting outbreak of listeriosis due to the contamination of a locally produced soft cheese, causing at least 122 cases, of which 31 were fatal [3]. As a consequence of this outbreak, the federal health authorities (Swiss Federal Office of Public Health, SFOPH) designated a National Reference Centre for Listeriosis (CNRL), one of the tasks of which is to collect and characterise L. monocytogenes isolates, primarily from humans, but also from animal, food and environmental samples taken in Switzerland. The CNRL operates in close cooperation with the clinical microbiology laboratories and the cantonal (regional) laboratories responsible for environmental surveillance and food safety. The report of culture-confirmed human cases of listeriosis to the SFOPH and the sending of isolates to the CNRL have been mandatory for laboratories in Switzerland since 1988.

Between 1990 and 2005, the annual number of culture confirmed cases of human listeriosis has varied between 14 (in 1990) and 70 (in 2005), corresponding to 0.14 and 0.9/100 000 inhabitants per year [4]. During this time period, the proportions of bacteraemia (40%), central nervous system (CNS) infections (40%), and materno-fetal infections (20%) remained relatively constant.

Methods

The laboratory surveillance consists of confirming the identification of the isolates to the species level and typing the L. monocytogenes isolates. Serotyping is carried out as a first step screening method using a commercial agglutination test (Denka Seiken, Tokyo, Japan) based on antibodies specifically reacting with somatic (O) and flagellar (H) antigens.

This step is completed with pulsed field gel electrophoresis (PFGE) if a cluster of isolates is observed, based on geographic consideration, multiple cases on a short period of time, or cluster of isolates with identical serotype. PFGE was done following the PulseNet protocol (PFGE after DNA digestion with the enzymes Apa I and Asc I) (http://www.pulsenet-europe.org).

Interviews with patients and analysis of milk products were conducted by the local food authorities, the regional chemistry laboratory (Service de la consommation, Neuchâtel). Patient interviews were carried out face to face or by phone by a specialist microbiologist from the regional laboratory.
**L. monocytogenes** in milk and milk products was detected according to the official methods in the Swiss Food Manual (http://www.bag.admin.ch/slmb/aktuell/d/56_Mikrobiologie.pdf). The method is based on an enrichment step, followed by plating on a selective agar and confirmation tests. The method for quantitative detection of **L. monocytogenes** on ALOA-agar was also used.

### Results

Within a period of 7 weeks in spring 2005, 10 human cases of listeriosis were diagnosed in a small area of northwest Switzerland (the canton of Neuchâtel, 150,000 inhabitants) by local physicians and clinical microbiologists. These 10 patients were admitted to three different hospitals [TABLE]. A single clinical microbiology laboratory serves these three hospitals and documented all 10 cases microbiologically. Four of the cases were in men (age range 70-72 years) and six were in women (two pregnant women, ranging in age between 23-26 years; and four non-pregnant women, aged 59-82 years). Clinical manifestations were bacteraemia confirmed. No CNS manifestation occurred. No other underlying disease or condition; three died within 30 days of admission to hospital, and both pregnancies ended in septic abortion. Two further adult patients living in neighbouring cantons were diagnosed with listeriosis with febrile gastrointestinal symptoms during the following two weeks.

All 12 isolates (10 invasive isolates and two isolates from stool samples) belonged to serotype 1/2a, which has been the most commonly reported serotype in human and food isolates in Switzerland since 1995 [4]. Before this outbreak, however, only 3/18 (17%) human isolates reported in 2005 were of serotype 1/2a [FIGURE], which led the outbreak investigators to suspect a common source. For this reason, and because of the unusually high number of human cases of listeriosis recorded in a short period of time, the physicians and clinical microbiologists involved suspected a common source of infection, and contacted the regional chemistry laboratory (Service de la consommation) in Neuchâtel, which also has responsibility for food safety in the area. On 4 June 2005, a microbiologist at this laboratory interviewed five patients face to face in hospital or at home, and conducted a telephone interview with a sixth patient. The interviews strongly suggested that a locally produced and distributed soft cheese known as a ‘tomme’ could be the origin of infections. Only one cheese factory in the region produced tomme cheese. Five samples of the suspected cheese were taken from the factory on 6 June and analysed immediately.

### Table

**L. monocytogenes** (serotype 1/2a) outbreak related to the consumption of Tomme cheese. Patient characteristics, Switzerland, 2005

<table>
<thead>
<tr>
<th>Case number</th>
<th>Sex</th>
<th>Age (years)</th>
<th>Date of isolation</th>
<th>Site</th>
<th>Hospital</th>
<th>Underlying disease or condition</th>
<th>Outcome</th>
<th>PFGE type (Apa I, Asc I)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>M</td>
<td>72</td>
<td>18 April 2005</td>
<td>Blood</td>
<td>1</td>
<td>Myeloma</td>
<td>Death</td>
<td>A</td>
</tr>
<tr>
<td>2</td>
<td>M</td>
<td>72</td>
<td>21 April 2005</td>
<td>Blood</td>
<td>2</td>
<td>Renal transplant</td>
<td>A</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>72</td>
<td>1 May 2005</td>
<td>Blood</td>
<td>1</td>
<td>Myeloma</td>
<td>Death</td>
<td>B</td>
</tr>
<tr>
<td>4</td>
<td>F</td>
<td>77</td>
<td>20 May 2005</td>
<td>Blood</td>
<td>2</td>
<td>Renal transplant</td>
<td>A</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>M</td>
<td>70</td>
<td>20 May 2005</td>
<td>Blood</td>
<td>3</td>
<td>Renal cancer</td>
<td>Death</td>
<td>B</td>
</tr>
<tr>
<td>6</td>
<td>F</td>
<td>80</td>
<td>27 May 2005</td>
<td>Blood</td>
<td>2</td>
<td>Renal dialysis</td>
<td>A</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>F</td>
<td>59</td>
<td>27 May 2005</td>
<td>Blood</td>
<td>1</td>
<td>Immunosuppressive drug</td>
<td>Death</td>
<td>A</td>
</tr>
<tr>
<td>8</td>
<td>F</td>
<td>26</td>
<td>31 May 2005</td>
<td>Blood</td>
<td>3</td>
<td>Septic abortion (22 w*)</td>
<td>A</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>F</td>
<td>23</td>
<td>1 June 2005</td>
<td>Placenta</td>
<td>1</td>
<td>Septic abortion (15 w*)</td>
<td>A</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>F</td>
<td>82</td>
<td>5 June 2005</td>
<td>Blood, stool</td>
<td>1</td>
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<td></td>
</tr>
<tr>
<td>11</td>
<td>M</td>
<td>53</td>
<td>7 June 2005</td>
<td>Stool</td>
<td>-</td>
<td>Febrile gastroenteritis</td>
<td>A</td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>F</td>
<td>47</td>
<td>17 June 2005</td>
<td>Stool</td>
<td>-</td>
<td>Febrile gastroenteritis</td>
<td>A</td>
<td></td>
</tr>
</tbody>
</table>

* Weeks of gestation

Swiss legislation decrees a limit for **L. monocytogenes** which is ‘not detectable in 25g’, and the regional laboratory analyses milk and milk products according to these criteria. In addition to that, the laboratory applied the method for quantitative detection of **L. monocytogenes** to gather information about the average **L. monocytogenes** counts in the suspected cheese. It should be noted that the method for quantitative detection of **L. monocytogenes** on ALOA-agar gives results faster than the presence-absence test in 25g of food, which takes between three and four days to complete. In the quantitative detection method, ALOA-agar plates are incubated at 37˚C for 24 to 48 hours. Using this method, one of the five cheese samples was found to be positive for **L. monocytogenes** the next day (7 June).
After enrichment, three of the five samples were found to be positive. In the following days, the laboratory analysed more samples which were delivered by consumers, and the public alert was raised. These samples were all found to be positive for *L. monocytogenes*.

After PFGE analysis with 2 restriction enzymes (Apa I and Asc I), the 12 human isolates, and food and environmental isolates, were found to be of two pulsovars [6]. Ten human isolates and 12 food isolates shared outbreak pulsovar A, and two human and 2 food isolates shared outbreak pulsovar B [TABLE]. Among 29 other serovar 1/2a *L. monocytogenes* isolates recovered during 2004 and 2005 that had been tested for pulsovars, none shared the pulsovars A or B. The PFGE subtyping clearly confirmed the epidemiological link between the incriminated cheese and 10 cases of invasive listeriosis. It is interesting to note that the 1983-87 Swiss outbreak was also caused by two different strains [3].

A large national retailer sold butter in this region that was produced in the incriminated cheese factory (this butter was only sold within the local area). When the outbreak was first reported, this company analysed unopened samples of butter in their own laboratories. Five out of 10 samples from two different lots were found to contain *L. monocytogenes* of serotype 1/2a in 25g, and therefore, according to legislation, could not be sold. The bacterial counts in the five samples were lower than 100 cfu/g, which is the detection limit of the method for quantitative analysis of *L. monocytogenes*. These findings were later confirmed by the regional laboratory. Up to 32,000 cfu/g of *L. monocytogenes* were found in the tomme cheese, a level of contamination significantly higher than that in the butter.

Since the interviews with the listeriosis patients clearly pointed to tomme cheese from a particular producer, risk management measures were taken before bacteriological results were available. Production of the suspected tomme was suspended, and cheeses sold under one particular brand name were recalled from the market, and a public alert and press information were released on 6 June. On 7 June, after 24 hours of incubation, one of five cheese samples showed presumptive colonies on ALOA-agar and thus confirmed the need for the measures that had been taken the day before. Furthermore, a legally binding order was issued to the management of the cheese factory by the national authorities, asking the factory to stop production and to perform environmental analyses in order to identify the weak points in the production process that had caused the outbreak. These investigations were done by microbiologists from Agroscope-Liebefeld (formerly the Swiss Dairy Research Station). It was demonstrated that *L. monocytogenes* was widespread throughout the facilities, but it was not possible to discover where the incriminated *L. monocytogenes* strains had originated. At the time of writing this paper, the cheese factory had not yet restarted production, although the required sanitary measures had been taken.

**Discussion**

Swiss food legislation decrees a microbiological criterion for *L. monocytogenes* in milk and milk products which is ‘not detectable in 25 g’. In the Neuchâtel outbreak, both tomme cheese and butter were found to exceed this limit and were therefore not acceptable under the current legislation. The EU regulation on microbiological criteria for foodstuffs, which will be incorporated into Swiss food legislation in the near future, differentiates between ready-to-eat food where *L. monocytogenes* can grow, and those foods where further growth is not likely. For the first group of foods, *L. monocytogenes* must be ‘absent from 25 g’, and for the second group, *L. monocytogenes* must not exceed 100 cfu/g. It is not clear to us how the EU regulation should be interpreted with regard to *L. monocytogenes* in butter. According to the findings of a Finnish study [5], it is possible for *L. monocytogenes* to grow in butter. For this reason, we think that butter also should comply with the requirement to have *L. monocytogenes* ‘absent from 25 g’.

The availability of a laboratory-based surveillance system with rapid typing, and the early raising of suspicion by local medical and microbiological staff, allowed rapid investigation of this outbreak and rapid recognition of the source. Considering the international distribution of many foods that may be a high risk for listeriosis infection, this illustrates the utility of an international surveillance network such as the one currently in development [7].

**References**

The first HIV/AIDS cases in Poland were diagnosed in the mid-1980s, and the outbreak in injecting drug users was first observed in 1989. For many years the HIV epidemic in Poland was driven by injecting drug use. In this study we examine the trends in the HIV/AIDS epidemic based on the surveillance data for 1999-2004. During this period, 3561 new HIV infections (annual rate of 15.4 per 1,000,000 inhabitants) were reported and 803 incident AIDS cases (incidence 3.5 per 1,000,000) were diagnosed. Both the annual number of newly detected HIV infections and the AIDS incidence showed a slight increasing trend. In particular, the vertically transmitted AIDS incidence increased from 0.46 in 1999-2000 to 0.91 per 1,000,000 children under 15 years in 2003-2004. Approximately 36% of AIDS patients aged 15 years or above had not been previously diagnosed with HIV. The annual number of the late presenters increased markedly between 1999 and 2004 and was higher amongst individuals infected through sexual transmission (51.0%) than those infected by injecting drug use (20.1%). Injecting drug users made up 78.6% of new HIV infections with known transmission route, but for 47.9% of all cases the route of transmission was not reported. In order to generate more accurate data, HIV surveillance must be enhanced. Nevertheless, there is clear evidence for implementation of a comprehensive programme of prevention of vertical transmission and encouraging more extensive HIV testing especially in the groups at risk for sexual transmission. An effort is needed to enhance HIV surveillance and prevention in the framework of programmes for STI.

A large proportion of the cases registered during the peak lacked sufficient identifying information and it is possible that some of these cases were registered again at a later time.

Highly active antiretroviral therapy (HAART) was first introduced in Poland in 1996, and in 1999 a special government programme coordinated by the National AIDS Centre was established, assuring general availability of free-of-charge therapy [3].

The aim of this study is to describe current trends in the epidemiological situation of the HIV/AIDS in Poland, based on the surveillance data from 1999-2004.

Methods

The surveillance system comprises reporting of newly diagnosed HIV infections as well as the incident AIDS cases. AIDS case notification is mandatory for all attending physicians, who complete standardised case report forms and send them to the regional public health departments (WSSE, Wojewódzki Stacje Sanitarno-Epidemiologiczne). Epidemiologists at the WSSE review the cases to check if the case definition criteria are met and collect additional information if necessary. Subsequently the WSSE forward the forms to the Department of Epidemiology of the National Institute of Hygiene.

The laboratories performing confirmatory HIV tests (immunoblot, PCR) report newly diagnosed HIV infections directly to the Department of Epidemiology.

With exception of two outbreaks – in Romanian children and, as mentioned above, in Polish IDUs - the rate of new HIV diagnoses in central Europe remains low and the epidemic is driven by sexual transmission.

Between 1985 and 2004, 9151 newly detected HIV infections and 1537 AIDS cases were registered in Poland. The AIDS incidence and the rate of detection of HIV infection cases after the early peak due to the epidemic among IDUs remained stable, with a consistent slow increase year on year [FIGURE 1].

**FIGURE 1**

Newly detected HIV infections and incident AIDS cases in Poland, 1986–2004

<table>
<thead>
<tr>
<th>Year</th>
<th>Newly diagnosed HIV infections</th>
<th>Incident AIDS cases</th>
<th>HIV infection cases excluding cases without full ID</th>
</tr>
</thead>
<tbody>
<tr>
<td>1986</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>1987</td>
<td>0</td>
<td>0</td>
<td>0</td>
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<td>1988</td>
<td>0</td>
<td>0</td>
<td>0</td>
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<tr>
<td>1989</td>
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<td>0</td>
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<td>1991</td>
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<td>1992</td>
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<td>1993</td>
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<td>1994</td>
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<td>1995</td>
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<td>1996</td>
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<td>1997</td>
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<td>1998</td>
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<tr>
<td>2003</td>
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</tr>
<tr>
<td>2004</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

A large proportion of the cases registered during the peak lacked sufficient identifying information and it is possible that some of these cases were registered again at a later time.
HIV/AIDS reports include personal identifiers: name (or only the initials), date of birth (or age), gender, address (or administrative region) and, recently, personal identification number, as well as the presumed mode of transmission. For cases of AIDS, data on indicator diseases and vital status are also required. Cases with known initials, date of birth and sex are considered to have the full identifier.

The Department of Epidemiology at NIH maintains a registry of HIV/AIDS cases. All newly reported cases are compared with the registry to avoid double registration; the case classification is once again validated.

The system registers all HIV infections diagnosed with definite methods and all confirmed AIDS cases according to the 1987 European case definition, taking into account the 1993 correction and the 1995 case definition for children [4, 5]. Each AIDS case must be linked to a record in the HIV registry.

In the present study data on newly detected HIV infection cases reported in 1999 – 2004 and on incident AIDS cases diagnosed during the same time period (reported until 31 March 2005) were included in the analysis. Reporting delays of over 3 months are uncommon.

### Results

#### HIV infection

During 1999 – 2004, 3561 newly detected HIV infections (annual rate 15.4 per 1 000 000) were reported through the routine surveillance system, 2584 (73.7%) in males and 923 (26.3%) in females [TABLE 1]. Injecting drug use was the most commonly presumed transmission route, accounting for 78.6% of infections with reported transmission route. Two other important routes of transmission included heterosexual contact (9.2%) and sexual contact between men (9.0%). In 47.9% of all HIV cases, however, the route of transmission was not reported.

HIV infections were detected in all regions in Poland, but the rate varied between the regions, with the lowest average annual rate of 3.1 per 1 000 000 inhabitants in Świętokrzyskie and the highest, in Dolnośląskie (34.7/1 000 000) and Warmińsko-Mazurskie (21.5/1 000 000) [FIGURE 2]. Among cases with reported transmission route, the proportion of IDU transmission was the highest in the two northeastern regions – 89.3% in Warmińsko-Mazurskie and 88.1% in

### Table 1

Number of newly detected HIV infections and incident AIDS cases, by sex, age groups and transmission route in Poland, 1999-2004

<table>
<thead>
<tr>
<th>Transmission route</th>
<th>MSM</th>
<th>IDU</th>
<th>HCA</th>
<th>Het</th>
<th>MtC</th>
<th>Unkn.</th>
<th>Total</th>
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</thead>
<tbody>
<tr>
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<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
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<td>107</td>
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<td>54</td>
</tr>
<tr>
<td>%Female</td>
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<td>Median age at diagnosis</td>
<td>41kys</td>
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<td>-</td>
<td>36yrs</td>
<td>2yrs</td>
<td>35yrs</td>
<td>34yrs</td>
</tr>
<tr>
<td>Total</td>
<td>118</td>
<td>433</td>
<td>2</td>
<td>140</td>
<td>28</td>
<td>82</td>
<td>803</td>
</tr>
</tbody>
</table>

MSM - men who have sex with men; IDU - injecting drug users; HCA - Health care associated; Het - heterosexual contact; MtC - mother to child; Unkn. - unknown

---

**Figure 2**

A. Regional variation of the rate of newly detected HIV infections and of the proportions of different transmission routes in Poland, 1999-2004

B. Administrative regions and major urban areas in Poland

**Administrative regions:**

1. Dolnośląskie
2. Kujawsko-Pomorskie
3. Lubelskie
4. Lubuskie
5. Łódzkie
6. Małopolskie
7. Mazowieckie
8. Opolskie
9. Podkarpackie
10. Pomeranian
11. Podlaskie
12. Świętokrzyskie
13. Warmińsko-Mazurskie
14. Wielkopolskie
15. Wielkopolskie
16. Złotowiejskie

**Average number of newly diagnosed HIV infections per 100 000 inhabitants:**

<table>
<thead>
<tr>
<th>Bracket</th>
<th>Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.65 to 3.47</td>
<td>(2)</td>
</tr>
<tr>
<td>1.24 to 2.15</td>
<td>(3)</td>
</tr>
<tr>
<td>0.98 to 1.24</td>
<td>(3)</td>
</tr>
<tr>
<td>0.66 to 0.98</td>
<td>(3)</td>
</tr>
<tr>
<td>0.31 to 0.66</td>
<td>(5)</td>
</tr>
</tbody>
</table>
Podlaskie - and the lowest in Mazowieckie (61.4%) and Malopolskie (62.5%). Heterosexual transmission was more common in Malopolskie (17.9%), Podlaskie and Swietokrzyskie (14.3%) and sexual transmission between men in Wielkopolskie (27.1%), Mazowieckie (22.9%) and Malopolskie (17.9%).

Overall, the median age at HIV diagnosis was 28 years and, excluding children of HIV infected mothers, and ranged from 26 years in the IDU to 34 years in people infected though sexual contact. Approximately 30% (n=1087) of the infected were under 25 years of age, including 60 children, who acquired the infection from their mothers [TABLE 1]. In recent years, however, the age distribution appears to have shifted towards older age groups [FIGURE 3].

**FIGURE 3**

Trend of age distribution of the newly detected HIV cases, Poland 1985–2004

A total of 803 AIDS cases were diagnosed during the study period, including 176 (21.9%) in females. The median age was 34 years, but the cases in MSM tended to be in older patients (median age 41 years) and those in IDUs, in younger patients (median age 32 years) [TABLE 1]. Approximately 36% of all cases, excluding children under 15 years, were diagnosed with AIDS within 3 months of HIV diagnosis (late presenters). Although overall AIDS incidence was stable over the years examined, the number of late presenters has recently increased sharply [FIGURE 4]. Late presenting cases, as compared to other cases, were more likely to be in people younger than 25 or older than 45 years, although mean age was comparable for the two groups (36.3 and 35.5 years for late presenters and others, respectively, p-value 0.283). The majority of the late presenters acquired their infection through sexual contact, while the IDUs predominated in the group of cases that were not late presenters [TABLE 2]. However, the transmission route was not reported for 20% of late presenters.

Between 1999 and 2004, 32 paediatric AIDS cases were reported, 28 transmitted vertically, one infected through blood transfusion and three for whom the route of transmission was not established. The AIDS incidence due to vertically transmitted HIV infection increased from 0.46 per 1 000 000 children younger than 15 in 1999 - 2000, to 0.64/1 000 000 in 2001 - 2002 and 0.91/1 000 000 in 2003 - 2004.

**TABLE 2**

AIDS cases diagnosed in 1999-2004, excluding children below 15 years. Comparison of characteristics of cases by the time of the HIV and AIDS diagnosis, Poland

**Discussion**

During 1999 – 2004 the registered rate of newly detected HIV infections continued to increase gradually. In contrast to other central European countries, the epidemic in Poland is unlikely to be fuelled by sexual transmission, although it exhibits marked regional variability. Given the currently increasing trends of heterosexual acquired HIV infections in the Newly Independent States of the Former Soviet Union, the possibility of augmented heterosexual transmission has become an important concern [6]. A study comparing early syphilis and gonorrhoea incidence in the eastern part of Poland in 1988/89 and 1996/97 demonstrated a significant increase of the percentage of STI patients in this region who acquired the diseases through sexual contact with a person from one of the neighbouring countries to the east [7]. However, in the period of time examined there was no evidence of increased homo- or heterosexual spread of the HIV epidemic in the eastern Poland. Conversely, the apparently injection-driven epidemic in northeast Poland near the Kaliningrad border suggests possible links with the Russian outbreak. However, because transmission route was not reported for a large proportion of these cases, these data must be interpreted with caution. Gender distribution of cases with unknown transmission route (72.4% males, 27.6% females) parallels that in IDUs (74.2% males, 25.8% females), indicating that injecting drugs could play an important role in the group with unreported transmission route. In comparison, the proportion of females among those infected heterosexually is higher (37.1%). However, those in the group with unknown transmission route were, on average, older than
Pregnant women are still not routinely being offered testing for HIV serostatus during the pregnancy [11]. Based on a study of over 25,000 transmission mainly occurs in women who did not know about their incidence of vertically transmitted AIDS in Poland continues to rise. The availability of the mother-to-child transmission prophylaxis since 1994, compartments such as injecting drug users. Furthermore, despite the Poland may be underestimated and not limited to specific population with reported transmission route) indicate that the HIV epidemic in Poland may be underestimated and not limited to specific population compartments such as injecting drug users. Furthermore, despite the availability of the mother-to-child transmission prophylaxis since 1994, incidence of vertically transmitted AIDS in Poland continues to rise. The transmission mainly occurs in women who did not know about their serostatus during the pregnancy [11]. Based on a study of over 25,000 newborns tested in 2001–2002 in the Mazowieckie region, between 100 and 200 seropositive women give birth each year in Poland [12]. Pregnant women are still not routinely being offered testing for HIV.

To conclude, in order to generate more accurate data, HIV surveillance must be enhanced by collecting detailed risk information. Even though further studies to guide prevention strategies are warranted, it is clear that implementation of a comprehensive programme of vertical transmission prophylaxis including voluntary testing of all pregnant women should be a priority. Moreover, there exists a need to increase access to and use of HIV testing by offering it more widely in accessible settings, or even by approving self-testing kits. Considering that the majority of late presenters were infected through sexual transmission, an effort is also needed to enhance collaboration between the HIV and STI surveillance and prevention programs.

References

Original Article
Surveillance report
G Asseva, P Petrov, I Ivanov, T Kantardjiev*

This article analyses the distribution of resistant salmonella and resistance mechanisms among the most frequently encountered serotypes in Bulgaria. Culture, biochemical tests and serotyping were used for identification. Screening for resistance to 14 antimicrobial agents with the standard Bauer-Kirby disk-diffusion method. The double disk-synergy method was used to determine production of extended-spectrum β-lactamases (ESBL). Transfer of genes coding for ESBLs with experimental conjugation. Specific primers were used for PCR detection of bla-CTX-M, bla-SHV and bla-TEM. 245 resistant salmonella strains were determined in our study, the majority originated from sporadic cases of human illness or asymptomatic infection and the remaining 23 were isolated from outbreaks. 79 producers of ESBL were detected: 5 S. Enteritidis, 1 S. Typhimurium, 9 S. Isangi and 62 S. Corvallis with types of enzymes: CTX-M3, TEM and SHV. Gene coding for extended-spectrum β-lactamases were successfully transferred into a recipient Escherichia coli C1 strain simultaneously with genes coding for resistance to aminoglycosides and sulphonamides (for bla-CTX-M3) and gene coding for resistance to aminoglycosides and chloramphenicol (for bla-SHV and bla-TEM). PCR amplification revealed bla-CTX-M3 genes in S. Enteritidis, and bla-SHV and bla-TEM in S. Corvallis. Salmonellae have revealed increasing resistance to all clinically important groups of antimicrobial agents. Bulgaria is the first country in the world where ESBL in serotype Corvallis has been reported. A wide diversity of resistance genes is found among the leading serotypes of salmonella causing human disease in Bulgaria.

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Keywords: ESBL-producing salmonellae, multidrug-resistant

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Introduction
The Bulgarian Ministry of Health has named salmonellosis one of the country’s priority communicable diseases, and healthcare providers are legally required to record and report cases of illness discovered in their regions. Surveillance of salmonellosis in Bulgaria is laboratory based. The network of microbiological laboratories functioning in the country performs the primary diagnosis and forwards both outbreaks and sporadic strains to the National Reference Laboratory for Enteric Pathogens for confirmation, serotyping and antimicrobial susceptibility testing. Since the second half of the 1990s Bulgaria participated in two international networks targeting surveillance of salmonellosis: Global Salm-Surv and Enter-net. National surveillance data on laboratory confirmed cases of human salmonellosis, including the total salmonella count and the total number of serotyped salmonellae, is reported annually to Global Salm-Surv. The databases enable us to follow the trends of these important infections at national and international level. Resistance to antimicrobial agents in enterobacteriaceae, including salmonella, is now an issue of international concern. The purpose of this paper is to analyse the distribution of resistant salmonella in Bulgaria and the resistance mechanisms among strains causing different forms of human illness: outbreaks, sporadic cases of disease and asymptomatic infection. In Bulgaria, as in many European countries, S. Enteritidis and S. Typhimurium are the first and second most common causative agents of human salmonellosis, respectively [1]. A shift in the position of S. Corvallis has been observed in our country since 1997 when it did not belong to the leading serotypes causing human salmonellosis. During the period under study, S. Corvallis was the third most common cause of salmonellosis in Bulgaria [FIGURE 1] [2].

**Figure 1**
Distribution of the top three Salmonella serotypes by number of laboratory confirmed cases, Bulgaria, 1999-2004

### Methods
A total of 6707 salmonella strains were isolated and reported by 28 Regional Inspectors for Prevention and Control of Public Health in Bulgaria between 1999 and 2004. These data were sent to Global Salm-Surv so that a country database could be established. 2123 (32%) of all salmonella isolates were sent to the National Reference Laboratory and were included in this study; 55 of them had caused outbreaks, and the remaining 2068 were from sporadic cases of salmonellosis or carrier state. Conventional microbiological methods: culture, biochemical tests, serotyping (BIO-RAD) have been performed for identification of strains. Screening for resistance to 14 antimicrobial agents (cefotaxime, Cefoxitin, carbencillin, cefazidime, cefuroxime, cephalothin, ampicillin, amoxicillin/ clavulanic acid, gentamicin, tetracycline, chloramphenicol, ciprofloxacin, nalidixic acid, trimethoprim/ sulfamethoxazole (Biomieuxes)) was done using standard Bauer-Kirby disk- diffusion method and screening for ESBL-production with the double disk synergy method [3]. All resistant strains were divided into two groups depending on their phenotypes:

1. resistant to < 4 antimicrobial agents
2. resistant to ≥ 4 antimicrobial agents.

### Results
A total of 245 resistant salmonella strains were found in our study. 222 (91%) originated from sporadic cases of salmonellosis or human carriers; 23 were obtained from outbreaks. Table 1 shows the distribution of resistant strains among the leading three salmonella serotypes causing human disease in Bulgaria for the period 1999-2004.

Characteristically, resistance increases in dynamics for S. Enteritidis, S. Typhimurium and S. Corvallis. Table 2 represents the distribution of resistant strains among the less frequently detected serotypes in Bulgaria for the period 1999-2004. Seventy nine of 245 strains (32%) produced extended-spectrum β-lactamases (ESBL): 5 S. Enteritidis, 1 S. Typhimurium, 9 S. Isangi and 62 S. Corvallis. The remaining 166 (68%) revealed resistance to ampicillin, carbenicillin, first and second generation cephalosporins, aminoglycosides, tetracycline, chloramphenicol, nalidixic acid, trimethoprim sulfamethoxazole alone and in combinations. ESBL producing salmonellae have demonstrated multidrug-resistance to more than seven antimicrobial agents, and were therefore classified into the group of microorganisms resistant to ≥ 4 antibiotics [TABLES 1 and 2]. This mechanism of resistance has been proved in strains originating from 6 regions situated in central and western Bulgaria. Bla genes coding for ESBL were successfully transferred into a recipient E. coli C1A strain simultaneously with the transfer of bla-CTX-M, bla-TEM and bla-SHV genes was studied with experimental conjugation using 14 salmonella strains as donors and an Escherichia coli C1A strain as a recipient. Transconjugates were selected on McConkey agar containing cefotaxime 10µg/ml and nalidixic acid 40 µg/ml. For PCR detection of bla genes the following primers were applied: ALA2/P2D for bla-CTX-M 5’ ATGTTAAAAAACGTCCG 3’ / S. typhimurium 5’ CTGATTTGCTGTAACAAAG 3’ [11], O55/O56 for bla-SHV 5’ TTATCTCCTGGTTACCCACCA 3’ / 5’ GATTGCTGATTTCGGCCG 3’ [12] and 5’-3’ ATGATGTTACCAATTTCCC AGCAATGTTAACAGTGAG for bla-TEM.

### Table 1
Resistant Salmonella among the top three serotypes detected in Bulgaria between 1999-2004

<table>
<thead>
<tr>
<th>Year</th>
<th>Total number of tested strains/ number of resistant strains (%)</th>
<th>Total number of resistant strains</th>
<th>Total number of ESBL-producing strains</th>
</tr>
</thead>
<tbody>
<tr>
<td>1999</td>
<td>230/ 31 (13.4)</td>
<td>25 (10.8)</td>
<td>6 (2.6)</td>
</tr>
<tr>
<td>2000</td>
<td>184/ 52 (28.2)</td>
<td>50 (27.1)</td>
<td>2 (1.1)</td>
</tr>
<tr>
<td>2001</td>
<td>72/ 24 (33.3)</td>
<td>23 (31.9)</td>
<td>1 (1.4)</td>
</tr>
<tr>
<td>2002</td>
<td>15/ 5 (33.3)</td>
<td>5 (33.3)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>2003</td>
<td>47/ 17 (36.2)</td>
<td>11 (23.4)</td>
<td>6 (12.8)</td>
</tr>
<tr>
<td>2004</td>
<td>24/ 10 (41.6)</td>
<td>9 (37.5)</td>
<td>1 (4.2)</td>
</tr>
</tbody>
</table>

| Total | 572/ 139 (24.3) | 123 (21.5) | 16 (2.7) |

### Table 2
Resistant Salmonella among the top three serotypes detected in Bulgaria between 1999-2004

<table>
<thead>
<tr>
<th>Year</th>
<th>Total number of tested strains/ number of resistant strains (%)</th>
<th>Total number of resistant strains</th>
<th>Total number of ESBL-producing strains</th>
</tr>
</thead>
<tbody>
<tr>
<td>1999</td>
<td>Not tested</td>
<td>Not tested</td>
<td>Not tested</td>
</tr>
<tr>
<td>2000</td>
<td>Not tested</td>
<td>Not tested</td>
<td>Not tested</td>
</tr>
<tr>
<td>2001</td>
<td>23/ 9 (39.1)</td>
<td>1 (4.3)</td>
<td>8 (34.7)</td>
</tr>
<tr>
<td>2002</td>
<td>38/ 38 (100)</td>
<td>0 (0)</td>
<td>38 (100)</td>
</tr>
<tr>
<td>2003</td>
<td>16/ 16 (100)</td>
<td>1 (6.3)</td>
<td>15 (93.7)</td>
</tr>
<tr>
<td>2004</td>
<td>6/ 5 (50)</td>
<td>2 (33.3)</td>
<td>1 (16.6)</td>
</tr>
</tbody>
</table>

| Total | 83/ 66 (79.5) | 4 (4.8) | 62 (74.6) |
Table 2

Distribution of resistant Salmonella among the less frequently detected serotypes in Bulgaria for the period 1999-2004

<table>
<thead>
<tr>
<th>Serotype</th>
<th>Number of tested strains</th>
<th>Number of resistant strains (% of total number of tested strains)</th>
<th>Resistant to &lt; 4 antibiotics (%)</th>
<th>Resistant to &gt; 4 antibiotics (%)</th>
<th>Number of ESBL-producing strains</th>
</tr>
</thead>
<tbody>
<tr>
<td>S. Isangi</td>
<td>19/15 (78.6)</td>
<td>1 (5.3)</td>
<td>1 (100)</td>
<td>0</td>
<td>9</td>
</tr>
<tr>
<td>S. Tshongue</td>
<td>1/1 (100)</td>
<td>0 (0)</td>
<td>1 (100)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>S. Gallinarum</td>
<td>3/3 (100)</td>
<td>3 (100)</td>
<td>0 (0)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>S. Gloucester</td>
<td>2/1 (50)</td>
<td>0 (0)</td>
<td>1 (50)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>S. Hadar</td>
<td>3/2 (66.7)</td>
<td>1 (33.3)</td>
<td>1 (33.3)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>S. Breda</td>
<td>1/1 (100)</td>
<td>0 (0)</td>
<td>1 (100)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>S. Blockley</td>
<td>3/2 (66.7)</td>
<td>2 (66.7)</td>
<td>0 (0)</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Discussion

In Bulgaria, the leading serotypes of non-typhoid salmonella have become resistant to most of clinically important antimicrobials. This major change had occurred during the last few years. Many countries consider resistance to nalidixic acid combined with retained susceptibility to ciprofloxacin to be the most typical mechanism for salmonella. Bacteria expressing such phenotypes have mutation in their chromosomal gyrA. In Bulgaria, this mechanism of resistance has been seen mainly in strains of S. Enteritidis. Despite the wide occurrence of ESBLs in Klebsiella pneumoniae, E.coli, Citrobacter, Proteus and other members of the family Enterobacteriaceae, they remained rare in salmonella until the second half of the 1990s. The number of salmonella serotypes expressing ESBL continues to increase [4-7]. The first ESBL-producing strains were detected in 1999 and belonged to serotypes S. Enteritidis and S. Typhimurium. Our study has revealed CTX-M-3 ESBL in S. Enteritidis. To date, 36 types of CTX-M β-lactamases are known. They are grouped into four clusters and CTX-M-3 enzymes have been classified into the first cluster of cephalosporins [8]. Few reports of blaCTX-M-3 harbouring S. Enteritidis are available in literature, for example in Poland in 2000 [9].

In comparison to other non-typhoid salmonellae, S. Typhimurium are regarded as more resistant[10]. In Bulgaria strains from this serotype expressed mainly resistance to ampicillin, carbenicillin, tetracycline and chloramphenicol, that could be explained with widely distributed plasmids in European countries. Only one of the S. Typhimurium strains in our collection expressed ESBL, but failed to hybridise with any of the primer pairs used in the study, though its phenotype was suggestive for TEM or SHV types of ESBLs [FIGURE 2B, lane 10]. This strain did not transfer resistance genes to the recipient E.coli C1A during the experimental conjugation possibly because of their chromosomal location. Selection and dissemination of multidrug-resistant S. Corvallis is a characteristic finding for our country and the majority of strains belonging to this serotype are ESBL producers. S. Corvallis is a rare serotype in Europe, but in Bulgaria, it has been the third most commonly reported causative agent of human infection after S. Enteritidis and S. Typhimurium since 1997. Our study has shown for the first time that the SHV type of ESBL is present in S. Corvallis. Salmonellae from serotype Isangi are characteristically resistant to multiple antimicrobial agents. Since their first isolation in Bulgaria in 1956, there has been little work on understanding the mechanisms of resistance among salmonellae from this serotype. Our study has revealed 9 CTX- M producing S. Isangi.

Figures

A. Bla-CTX-M3 in S. Enteritidis and transconjugants
B. Bla-SHV in S. Corvallis and transconjugants


References

In 2001 Germany implemented a new electronic reporting system for surveillance of notifiable infectious diseases (SurvNet@RKI). The system is currently being used in all 431 local health departments (LHD), the 16 state health departments (SHD) and the Robert Koch-Institut (RKI), the national agency for infectious disease epidemiology. The SurvNet@RKI software is written in MS Access 97 and Visual Basic and it supports MS Access as well as MS SQL Server database management systems as a back-end. The database is designed as a distributed, dynamic database for 73 reporting categories with more than 600 fields and about 7000 predefined entry values. An integrated version management system documents deletion, undeletion, completion and correction of cases at any time and entry level and allows reproduction of previously conducted queries. Integrated algorithms and help functions support data quality and the application of case definitions. RKI makes the system available to all LHDs and SHDs free of charge. RKI receives an average of 300,000 case reports and 6240 outbreak reports per year through this system. A public web-based query interface, SurvStat@RKI, assures extensive and timely publication of the data. During the 5 years that SurvNet@RKI has been running in all LHDs and SHDs in Germany it has coped well with a complex federal structure which makes this system particularly attractive to multinational surveillance networks. The system is currently being migrated to Microsoft C#/NET and transport formats in XML. Based on our experiences, we provide recommendations for the design and implementation of national or international electronic surveillance systems.

Methods

Background and requirements

Germany is a federal republic with 16 states (Bundesländer) and 439 counties (Stadt-/Landkreise). Typically, there is one local health department (LHD) per county, responsible for managing single cases and outbreaks of infectious diseases and carrying out necessary prevention and control activities. The IfSG defines 47 pathogens and 14 diseases that laboratories and clinicians, respectively, have to notify to the local health department. LHD complete and verify the case information based on national case definitions. These cases are then transmitted on a single case basis to the state health departments (SHD) and from there to the Robert Koch-Institut (RKI), the central national agency for infectious disease epidemiology. A requirement analysis revealed the need for an electronic reporting system with the following functional and non-functional features:

The system capacity needed to be sufficient for over 300,000 reported cases per year with 25 to 60 variables per case entered by 431 LHDs throughout the country. The system needed to take issues of data security of privacy-related patient data as well as specific additional requirements of individual states into account. For economic reasons the software had to run on common hardware without the need for additional software licenses and expensive back-end systems. As permanent internet connection was not available in all LHDs, the system needed to be operable offline as well. The system should incorporate reporting of complex outbreaks and be flexible enough to adapt quickly to unexpected changes caused by new emerging diseases (e.g., SARS).

In July 2000 the two legislative houses of representatives in Germany (Bundestag and Bundesrat) ratified the IfSG to be enacted by 1 January 2001. Within 6 months the RKI developed the electronic reporting system for the national surveillance system.

Software design

The architecture of the system was designed inhouse at the RKI. However, a major part of the programming was done by an external IT company. The newly developed system was called SurvNet@RKI.

The data flow is depicted in the figure. The front-end of SurvNet@RKI is written in MS Access 97 and Visual Basic. Depending on the data volume it supports MS Access as well as MS SQL Server database management systems as a back-end. Adding or removing reporting categories, fields or allowed values do not require changes to the programme structure, which is based on the fractal concept of

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Keywords: disease notification, electronic reporting, infectious diseases, surveillance
picoware GmbH. SurvNet@RKI allows the reproduction of previously conducted queries and analyses by means of an integrated version management system: Any updates of a data record (case or outbreak) result in the creation of a complete new record in the database that is marked as valid beginning at the time when the record was created. The old record's validity period ends at that time. As shown above the data replication is organised by the transmission of transport files in a format specified by the RKI. The transmission format is text-based and allows the representation of complex data with possibly multiple nominations of a field. Online help functions provide additional information for the user (for example, the disease-specific case definition). Integrated algorithms that follow the national case definitions assure that case records are exported only if the case confirmation criteria are met.

Deletion is integrated into the transport process by activation of a marker which makes retrieval of previously deleted records possible.

**Data base design and management**

The database is designed as a dynamic, relational database that currently consists of 73 reporting categories with more than 600 fields and about 7000 predefined entry values in look-up fields. All criteria formulated in the national case definitions are integrated into the data entry forms in order to facilitate application of and compliance with case definitions [5].

Furthermore, each record representing a case of an infectious disease can belong to one or more groups of cases representing an outbreak. The version management described above is also applied to outbreak records.

**Results**

**Software design**

The RKI provided the commercial software manufacturers with the final technical specifications for electronic case reporting in October 2000 and released its own software programme, SurvNet@RKI, in December 2000, free of charge. The new system was implemented nationwide on 1 January 2001. Within a few weeks, almost all LHDs were reporting at least weekly through the new system [2]. All state health departments use SurvNet@RKI. Among the 431 LHDs in 2005, 112 (26%) use SurvNet@RKI while 319 (74%) use one of five different commercially available software programmes for public health administration which include a case reporting module based on the specifications published by RKI. Public health nurses at the LHD enter the data into the reporting software and complete the records according to findings of subsequent investigations. When outbreaks occur, the LHD (or the SHD or the RKI) creates an electronic outbreak record, which groups the affected case reports and holds additional data regarding the outbreak, such as modes of transmission and evidence for this information. At least once a week each LHD creates a transport file containing all changes since the last export. Those data are automatically extracted by the system. Any information subject to data privacy remains physically in the database of the LHD. The transport file is sent via email to the SHD, where the data is imported into SurvNet@RKI, which in turn generates a confirmation file that is sent back to the LHD, also via email. In all SHD and in those LHD that use SurvNet@RKI changes and additions in field definitions and database structure are usually executed within one month after publication of the new specifications. New versions of SurvNet@RKI are fully downward compatible, which ensures that data generated by older software versions can still be imported and handled.

**Data base design and management**

The RKI receives an average of 300 000 reported cases per year. Thirty six per cent of the case reports are completed or corrected during the investigation process and are therefore transmitted in two or more different versions, which are all retrievable in the database. This results in a total of 490 000 datasets sent to the RKI each year. Based on the complex record versioning system, datasets are never frozen at any given deadline but can be continuously corrected, completed, deleted and undeleted if necessary. Historical case counts can therefore be performed for any state of knowledge in the past, which facilitates the generation of epidemiological reports and comparison of data.

Eight per cent of the fields in SurvNet@RKI, such as reporting week and year, are mandatory fields, and must be filled in or the record cannot be saved. About 10% of the fields undergo an integrated plausibility algorithm (for example, the order of the timestamps for date of birth, onset of illness and date of diagnosis). This will generate error messages when data has not been entered or is in conflict with entries in other fields. Case reports of disease with a yearly incidence of approximately less than 1 case per 100 000 population undergo a manual quality control procedure at the RKI before they are released for publication;

**Figure**

Data flow in the German computerized reporting system
these cases made up 0.89% of the mean number of yearly reports (n = 1 212 482) from 2001-2004. Most data quality indicators have improved significantly over the past four years, but show variations depending on the state where the data is generated and the kind of software used to enter and manage the data at the LHD [2,4,6].

**Development effort**

The estimated cost for the development of the initial software prototype adds up to one year full time equivalent (FTE) for an IT scientist, one year full time equivalent for a medical epidemiologist and EURO 50 000 worth of external programming work. Furthermore, an estimated amount of 1 FTE for an IT scientist and 0.5 FTE medical epidemiologist in addition to EURO 60 000 for programming alone externally been invested each year for maintenance and further improvement of the system. This comes to a total of approximately EURO 170 000 for the initial development, plus EURO 150 000 per year for improvements and maintenance. It does not include the actual epidemiological work for data quality control, system evaluation, scientific interpretation of the data, and the training of external users of the system.

**Data release and publication**

The national surveillance data collected at RKI are published periodically [6] or whenever required by RKI staff or external scientists. In order to improve data quality, implausibilities are fed back to the SHD, and are forwarded from there to the appropriate LHD requesting validation or correction. SurvStat@RKI, a web-based query interface, allows interested users to perform analyses on the national data [7]. Each spring following the reporting year, RKI releases an annual epidemiological report of over 170 pages. Germany contributes more case reports than any other country to the European Basic Surveillance Network, which is facilitated by the ability of SurvNet@RKI to automatically translate the German raw data to the European data formats [8]. RKI also reports surveillance data electronically to the World Health Organization and to various dedicated surveillance networks of the EU.

**Outbreaks detected**

Interlinked with the reported individual cases, RKI receives an average of 6240 outbreak reports per year, which generally have been primarily identified and investigated by the LHD. On average, 2047 (33%) of these outbreaks have five or more cases [9]. In addition to assessing outbreaks detected at the LHD level, SurvNet@RKI has also been able to report outbreaks and clusters that were not identifiable at the SHD or LHD level because of their rather diffuse geographical distribution. Examples for such outbreaks are an outbreak of *Salmonella* Agona from contaminated aniseed [10], an international outbreak of *Salmonella* Oranienburg due to German chocolate [11] and a large outbreak of hepatitis A among German tourists returning from a hotel in Egypt [12].

In 2003 SurvNet@RKI has also been adopted for internal use in the German Armed Forces, contributing to a better information exchange between civil and military health departments as shown in a large outbreak of epidemic conjunctivitis [13].

**Discussion**

SurvNet@RKI has proved to be a powerful reporting system for cases and outbreaks of notifiable infectious diseases. Many national surveillance systems rely on or are moving towards electronic reporting systems (such as NEDSS in the United States [14], CIDR in Ireland [15], and SMINet in Sweden [16]). In comparison to these systems SurvNet@RKI provides some features of database and communication architecture that make the system particularly useful for surveillance networks of multiple states or countries and for environments in which requirements of data security and limitations to data sharing usually create major obstacles.

SurvNet@RKI addresses these challenges by using a physically distributed database characterised by a highly standardised core database and variable branch subsets. Another remarkable, and to our knowledge unique, feature is the tight integration of case reporting and outbreak reporting.

During the five years that the system has been running in all Germany’s LHDs and SHDs, it has coped well with a complex federal structure, which generally complicates or even impedes efficient information exchange between administrative levels.

We believe a key to the success of SurvNet@RKI was the very strong cooperation of epidemiologists from LHDs, SHDs and RKI, the in-house IT staff and the external company. The costs have been kept low.

However, we also experienced difficulties in implementing necessary changes rapidly throughout the country, particularly because manufacturers of commercial software at the LHD level took a long time to implement the changes, and in some cases were unable to implement the specifications at all. This puts LHDs who use such software programmes at a significant disadvantage, because the majority of the system changes aim to reduce the workload at the LHD level and to avoid data entry errors.

The use of MS Access 97 with Visual Basic programming proved to be an effective basis for finalising a stable prototype within a very short time. However, now after approximately five years of experience, recurring changes and amendments have resulted in a complexity of the system that is becoming hard to maintain with the current platform. For similar projects we recommend the use of professional development environments, object-oriented approaches, and data exchange technologies that are better at supporting team development, code reuse and change management.

We are currently migrating SurvNet@RKI to a new platform that better meets those requirements. It will be re-implemented in Microsoft C#/.NET. The former transport file format specification will be replaced by an XML schema. This allows, for instance, the manufacturers of third-party products to test their export files against the specification eliminating a frequent error source. The user interface will be multilingual.

In the framework of a federal government initiative (BundOnline 2005) to foster e-government solutions, we intend to develop an interface for the most commonly used laboratory software systems in order to enable laboratories to report automatically in electronic format to the respective LHDs.

**Recommendations**

Based on our findings and experience in designing and implementing SurvNet@RKI, we have come up with the following recommendations for future developments of multistate electronic reporting systems:

- Adhere to the best practices in software engineering. We recommend following an agile development process to keep costs low. Staff the team with both IT specialists and epidemiologists.
- The number of fields per case needs to be kept to a minimum. In contrast to the general tendency to expand the amount of data, revisions of the system should always aim to reduce complexity of the database. The more experience available on the quality of the incoming data and on its actual contribution to epidemiological conclusions, the easier it will be to keep the database simple.
- Drop-down menus presenting the choice of data field entries need to be formulated in clear, concise language that can be understood without advanced medical knowledge.
- Transport and interface formats should be based on XML.
- Software development should not be completely outsourced from the institution that will be in charge of the system. First, the epidemiological expertise needs to be included into the process from the beginning on, which is more efficiently done if the software design is done inhouse as well. Second, maintenance and improvement of the system requires inhouse IT expertise, otherwise sustainability is at risk or may become costly.
- Cooperation with multiple peripheral software manufacturers.
Electronic systems for communicable diseases surveillance enhance quality by simplifying reporting, improving completeness, and increasing timeliness.

In this article, we outline the ideas and technologies behind SmiNet-2, a new comprehensive regional/national system for communicable disease surveillance in Sweden. The system allows for reporting from physicians (web form) and laboratories (direct from lab data system) over the internet. Using a unique personal identification number, SmiNet-2 automatically merges clinical and laboratory notifications to case records. Privileged users, at national and county level, work against a common central server containing all notifications and case records. In addition, SmiNet-2 has separate county servers with tools for outbreak investigations, contact tracing and case management.

SmiNet-2 was first used in September 2004. Individual counties receive up to 90% of all notifications electronically. In its first year, SmiNet-2 received 54,980 clinical notifications and 32,765 laboratory notifications, which generated 58,891 case records. Since most clinicians in Sweden have easy access to the internet, a general web-based reporting has been feasible, and it is anticipated that within a few years all reporting to SmiNet-2 will be over the internet. In this context, some of the major advantages of SmiNet-2 when compared with other systems is timeliness in the dataflow (up to national level), the full integration of clinical and laboratory notifications, and the capability to handle more than 50 diseases with tailor-made notification forms within one single system.

**References**


**Keywords:** epidemiology, communicable diseases, infectious diseases, surveillance

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Introduction

Communicable disease surveillance is an ongoing process involving the systematic collection, analysis, interpretation and dissemination of health data. It aims to detect outbreaks early on, to monitor and analyse trends, and define public health priorities in order to reduce morbidity and mortality and achieve improved health [1-3]. A well-designed and functional surveillance system is fundamental for providing the necessary information for appropriate and timely action and response. In recent years, electronic reporting has become increasingly widespread, incorporating internet-based data entry for the notifying physician and/or the county/state health department, and automated input of electronic laboratory results [4,5]. Electronic systems may enhance the quality of the system by simplifying the reporting for the end users, improving the sensitivity (completeness of reporting), and the timeliness within the system, from event to action [6-13].

In Sweden, a national electronic surveillance system (SmiNet-1) has been in place since 1997. For security reasons, SmiNet-1 was built on a Lotus Notes platform, with local servers in each county and a central server at the SMI. The notification reports were manually entered at the CMO offices (clinical notifications) and at the SMI (laboratory notifications). Some major laboratories had export routines for exporting data directly from the laboratory computer systems to SmiNet-1. For clinical notifications, fields for all information on the standard report forms were at hand, while for the laboratory notifications, more specific information, such as antimicrobial susceptibility and genetic typing information could only be reported as non-standardised information in free text fields. Each night, the central and local Notes servers exchanged information on the recently entered notification information. For further data cleaning and analysis, an SQL-database (EpiArk) was used at the SMI and a stand-alone Lotus Notes application was used in many of the CMO offices. As there was no communication between EpiArk and the local databases, changes and updates made by CMO users were not available for SMI users and vice versa. Furthermore, patients with chronic infections such as HIV or hepatitis C could have separate case records in several of the local databases, but only one in the central database. The incidence for these infections from the county statistics were therefore higher than the county-level statistics submitted from the SMI. After a technical revision of SmiNet-1 in 2001, the inherent weaknesses in the system and outmoded IT solutions prompted the development of an entirely new system (SmiNet2), built using the experience and insight gained from SmiNet-1.

The reporting system

The Swedish Communicable Disease Act [14] regulates the reporting of 59 statutory notifiable infectious diseases. Diseases are notified in parallel by both the patient's physician (clinical notification) and the laboratory that has diagnosed the causative agent (laboratory notification) to the 21 county medical officers (CMOs) and to the Department of Epidemiology, Swedish Institute for Infectious Disease Control (EPI/SMI). The clinical notifications must contain detailed epidemiological information and the laboratory notifications, the relevant microbiological information. With the exception of sexually transmitted infections, all notifications are made using full patient identity, including a unique personal identification number that is issued to all Swedish residents. This number is used to link clinical and laboratory notifications on the same patient and disease episode.

Data-entry close to the source

One of the important ideas behind SmiNet-2 is for data entry to be made as close to the source as possible. Since all hospitals and health centres and almost all private physicians in Sweden have internet access, data-entry over the internet is the preferred mode of clinical notification. The system also allows detailed data to be imported from the microbiological laboratories’ computer systems without the need for manual data entry.

User groups

There are four groups of users in SmiNet-2, listed below. All users working within the same database.

Clinicians: There are about 30 000 clinicians in Sweden, working in approximately 5000 healthcare units (hospitals, health centres and private clinics). The clinician reports to the system using either a web interface or a paper form. The clinician may use the web interface to fill in the form and print it out before sending. The physician does not have access to any data within the system (one-way communication only).

Laboratories: There are about 50 routine microbiological laboratories in Sweden, including the reference laboratories. A laboratory has a choice of three reporting methods: through a direct connection from the laboratory data system to the SmiNet-2 using a web service; manually using a web interface; or using a paper form. All communication for the laboratories is one-way.

CMOs: The CMO has the overall responsibility for communicable disease surveillance and control within his county. The SmiNet-2 users at the CMO offices use a Java client for a two-way communication with SmiNet-2, for example, to enter additional information from clinicians and laboratories, to work with outbreak investigations and to get IT support when performing contact tracing.

EPI/SMI: The EPI/SMI is responsible for national surveillance of communicable diseases. The EPI/SMI staff use a Java client for a two-way communication with the system, for example, data cleaning and analysis.

Basic entities in the system

There are six basic entities in SmiNet-2 (listed below and illustrated in Figure 1).

Notification: A notification (clinical or laboratory) contains both mandatory information (for example, patient ID, diagnosis and date of reporting) and optional information (such as country and date of infection). The EPI/SMI has access to all notifications, while the CMOs have access only to notifications reported from their county.

Figure 1

Illustration of the relationship between the basic units

Patient record

| Case record |
| Laboratory notification |
| Note |
| Contact-tracing record |
| Note |

Investigation record

| Case record |
| Laboratory notification |
| Note |

Note

Data-entry close to the source

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Note

Case record: A case record in SmiNet-2 summarises information from all notifications for the same individual related to a specific disease and within a specified timeframe (defined for all diseases). Each case record can be associated with a patient record and/or an investigation record (see below). The EPI/SMI has access to all case records, while the CMOs have access only to the case records for which they have received a notification. If a patient has moved between counties, and been notified with the same infection (typically chronic infections such as hepatitis C) in more than one county, several CMOs may access the...
same case record. The case records form the basis for statistics, and a case record may therefore be active in only one county at a time.

**Patient record:** A patient record represents a unique individual within the system. Each patient record is linked to his or her case records and contact tracing records. The patient record contains personal information, such as contact details and specific instructions given to the patient by the clinician, and can only be accessed by the CMO who created it.

**Investigation record:** An investigation record is used to gather and analyse information from outbreaks and other health events. Each investigation record can be linked to the case records associated with the outbreak. The investigation record can only be accessed by the CMO who created it.

**Contact tracing record:** SmiNet-2 provides the tools necessary for follow up of contact tracing at the CMO offices. Each contact tracing record is linked to a patient and can only be accessed by the CMO who created it.

**Note:** A note is created for recording an administrative event, such as a phone call, a letter or a decision. Each CMO can create letter templates to write standard letters, such as letters containing instructions to a patient. Each note may be linked to one or more case record, patient record, investigation record or contact tracing record. A note can only be accessed by the CMO who created it.

**Software and hardware**

SmiNet-2 is written in Java 2 Standard Edition (J2SE). The web module uses Java 2 Enterprise Edition (J2EE) IntelliJ IDEA (version 3.0.5) was used to develop the system. Two database servers are used. The CMO local databases use a MySql database server (version 4.0.20) and the central server databases use a Microsoft Server 2000 (version 8.0.0.194). Two Java Database Connectivity (JDBC) are used: a MySql JDBC Connector (version 3.0.6) and an i-net Merlia 2000 (version 1.03). Apache (version 2.0.46) is used as a web server, in conjunction with a Jakarta Tomcat (version 4.1.30) as application server and Apache Axis (version 1.1) for web services. Communication between the clients and the server are achieved using Java Remote Method Invocation (Java RMI) and the distribution of the clients is done using Java Web Start (JWS). Java Runtime Environment (JRE) version 1.4.2 or higher are required to run either a client or a server. OpenSSL (version 0.9.7a) is used for client server encryption.

**System architecture**

Figure 2 illustrates SmiNet-2’s system architecture.

**Server:** There are 22 different servers within the system, one central server (at SMI) and 21 local county servers. The central SmiNet server contains a number of databases. Each CMO has his or her own local SmiNet server, containing a local database with information that can only be accessed by the CMO (patient records, investigation records, contact tracing records and notes).

**Central databases:** The central server contains two databases: OrgArk (originals archive) and EpiArk (epidemiological archive). For legal reasons, OrgArk contains all notifications reported to SmiNet-2 in their original form. EpiArk contains all approved notifications (clinical and laboratory) and the corresponding case records. Either the CMO or the SMI must approve a notification to create it in EpiArk, and both must approve a notification to allow further processing. In EpiArk, a notification or a case record may be modified or supplemented (with full logs of all changes made, by whom and when). The central server also has separate administrative databases, for example, for user information and system logging.

**Local databases:** The local county databases contain patient records, investigation records, contact tracing records and notes. The information in these databases can only be accessed by the respective CMO (and his/her authorised staff). The local databases also store relational information, for example, to link a contact tracing record to a patient record, or a note to an investigation record.

**Clients:** Each group of SmiNet-2 users has its own specific way of interacting with the system. The clinicians log into the reporting form at the SmiNet website (http://www.sminet.se) using their workplace’s specific healthcare unit code, issued by the CMO.

A laboratory with export routines to SmiNet-2 in place in its laboratory data system creates an export file in a specified XML format, which is transferred to SmiNet-2 through a web service. If a laboratory cannot make the proper system adjustments, the notifications may be entered and sent manually using a web client.

The CMOs and EPI/SMI have Java clients to communicate with the central and local servers.

**Data security and safeguard of personal integrity**

The two-way communications between SmiNet-2 and the Java clients of EPI/SMI and the CMOs run over a private internet...
Survey reports

(wide area network (WAN) with restricted access) used by the Swedish healthcare services. All other clients work over the internet, but the functionality is limited to reporting (one-way communication). Login is required for all users, and all communication between client and server is protected by a strong SSL (Secure Socket Layer) encryption (168 bit 3DES) [15].

Only authorised staff at the CMO offices and at EPI/SMI, with pre-installed Java clients, can access the central database, and authentication is required. All staff with access to SmiNet-2 work under the same strict confidentiality rules that apply for direct patient contacts within the healthcare sector. Under the Swedish Secrecy Act [16], access to any healthcare related data is restricted to staff who need this data to fulfill their duties, and it should be directly related to the purpose for which the data were collected.

Introduction of SmiNet-2

SmiNet-2 was first used in September 2004, when two pilot counties and EPI/SMI began to use the system. The final county is scheduled to enter the system by mid-2006. The two pilot counties now receive between 80% and 90% of all notifications electronically. In its first year, SmiNet-2 received 54,980 clinical notifications (12% submitted electronically) reported by 1935 healthcare units and 32,765 laboratory notifications (78% submitted electronically) reported by 47 laboratories, which generated 58,891 case records. All case records from 1997–2005 stored in SmiNet-1 (approximately 390,000) have been migrated to SmiNet-2, and when the last county enters SmiNet-2, the old system will be closed down. Information on tuberculosis and HIV infections that have previously been stored in separate databases will also be fully integrated into SmiNet-2 during 2006.

Data output

EPI/SMI supplies web statistics on the communicable disease situation in Sweden, as tables, graphs and GIS maps for the SMI website [17].

Discussion

Other countries have implemented electronic web-based reporting mechanisms in their national surveillance, the Netherlands (Infectious Disease Surveillance Information System - ISIS) [7,18], the Republic of Ireland (Computerised Infectious Disease Reporting – CIDR) [19], and the United States (National Electronic Disease Surveillance System – NEDSS) [20,21]. In NEDSS, different states are using various computerised and web-based technologies. RODS (Real Time Outbreak and Disease Surveillance) is one of the latest technologies, and is increasingly being applied [5,22,23].

Each of these systems has its own profile and history, and any comparison between systems must take the local context into consideration. Sweden benefits from being a small country with a largely uniform organisation of health services, universal use of personal identification numbers, and a tradition of quality and comprehensiveness in reporting [9,10]. Since almost all clinicians have easy access to the internet, a general web-based reporting has been feasible, and it is anticipated that within a few years, almost all infectious disease reporting will be over the internet. In this context, some major advantages of SmiNet-2, compared to the old SmiNet-1 system and the systems in most other countries, include timeliness in the dataflow (up to national level), the full integration of clinical and laboratory notifications, and the capability to handle more than 50 diseases with tailor-made notification forms within one single system. The obvious gain in timeliness is due to direct entry of data at the source and therefore no delay in the mail process and data entry at the receiving end. We are planning a more formal evaluation in 2007, making use of the same methodology previously utilised when evaluating SmiNet-1 to more precisely quantify this gain [9,10]. Another unique feature of SmiNet-2, to our knowledge, is that it has built-in administrative databases and tools for the daily public health work such as outbreak investigations and contact tracing.

Direct links from the patient record systems of the health centres to SmiNet-2 will be an important function and will decrease the workload of the reporting physician, increase data quality and obtain timelier data. This modification has been considered, but an obstacle has been the wide range of different patient record systems. SmiNet-2 is currently being prepared to directly import data from these systems, using the same technology as for communicating with the laboratories, but export routines in the patient record systems need to be implemented by patient record system manufacturers.

A current weakness of SmiNet-2, compared with some other web-based systems, such as the German SurvStat@RKI system [24], is limitations on the output side. The system includes a number of data retrieval tools and reporting forms for the privileged users with Java clients at the EPI/SMI and the CMO offices, and these tools will be further developed in the near future. However, for the non-privileged users, with no direct access to the system, data is presented on the SMI website in static format only. Despite a number of output options (maps, graphs and tables), there is currently no possibility of retrieving data using one's own search criteria [17]. A priority for the future is therefore to make the output functions also on the website more diverse and user friendly.

As yet, there is no alert system integrated in SmiNet-2. In order to optimise the capacity of the system to detect outbreaks and other unexpected events, data need to be timely and algorithms need to be implemented to detect clusters of patients in time and space. To prepare SmiNet-2 for an early warning system, a study comparing three widely used algorithms have been conducted [25].

In 2005, the new European Centre for Disease Prevention and Control (ECDC) became operational [26]. One of the main tasks of the centre is to coordinate all European level surveillance activity on communicable diseases and to host the databases for this purpose [27], and the ECDC will need to evaluate closely the existing electronic surveillance networks in Europe and draw on the best practices available. The experiences from Sweden and those other countries that have recently been developing modern electronic surveillance systems will provide a good basis for this important future work.

Acknowledgements

Author KE is former Deputy State Epidemiologist for Sweden and was working at the Department of Epidemiology, SMI (EPI/SMI), during the main part of the development phase of SmiNet-2. The SmiNet-2 system is the result of the joint work of many people, especially the staff of EPI/SMI, but also the user groups of stakeholders.

References


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Eighty six new clusters were detected, 45% of which would not be associated with an accommodation site, EWGLINET initiates and ensures that international standards are adhered to. The history of EWGLINET, the number of cases reported to national surveillance schemes in 37 countries [1] (including hospital-acquired and community-acquired cases, as well as travel-associated cases), compared with only 242 in 1993 from 19 countries. This increase in numbers can be attributed to an increasing awareness of the disease, a rise in the number of contributing countries, and strengthening of national and international surveillance systems. Of the total cases recorded in 2004, 396 (8.6%) died.

This paper provides results and commentary on cases of travel-associated legionnaires’ disease with onset in 2004 reported to EWGLINET. The number of cases reported to national surveillance schemes across Europe has been increasing. In 2004, 4588 cases were recorded in 35 countries [1] (including hospital-acquired and community-acquired cases, as well as travel-associated cases), compared with only 242 in 1993 from 19 countries. This increase in numbers can be attributed to an increasing awareness of the disease, a rise in the number of contributing countries, and strengthening of national and international surveillance systems. Of the total cases recorded in 2004, 396 (8.6%) died.

This paper provides results and commentary on cases of travel-associated legionnaires’ disease with onset in 2004 reported to EWGLINET.

Methods
The addition of Andorra during 2004 brought the number of collaborators participating in EWGLINET to 59, representing 51 collaborating centres in 37 countries [FIGURE 1] which report all travel-associated cases fulfilling EWGLINET’s case definitions and detected by their national surveillance systems to the European database. Some countries host more than one collaborating centre. Collaborators are encouraged to report cases in people who travel within their own countries as well as those who travel abroad, and an increasing number of them are doing so.

Standard case definitions have been agreed by the collaborating countries in EWGLINET and are used for the purposes of international surveillance. A single case is defined as a person who, in the two to three days following their return from travel, develops symptoms of legionnaires’ disease. A single case is defined as a person who, in the two to three days following their return from travel, develops symptoms of legionnaires’ disease. When a cluster of cases is suspected to be associated with an accommodation site, EWGLINET initiates and monitors immediate control measures and investigations at the site, and ensures that international standards are adhered to. The history of EWGLINET and the number of cases reported to national surveillance schemes in 37 countries [1] (including hospital-acquired and community-acquired cases, as well as travel-associated cases), compared with only 242 in 1993 from 19 countries. This increase in numbers can be attributed to an increasing awareness of the disease, a rise in the number of contributing countries, and strengthening of national and international surveillance systems. Of the total cases recorded in 2004, 396 (8.6%) died.

This paper provides results and commentary on cases of travel-associated legionnaires’ disease with onset in 2004 reported to EWGLINET.
A cluster of travel-associated legionnaires' disease is defined as two or more cases in people who stayed at or visited the same accommodation site in the two to ten days before onset of illness and where onset is within the same two-year period [2].

Cases are initially reported to their national surveillance schemes, which gather all relevant details on the case, such as information on microbiological diagnoses and travel history, and then report them to the EWGLINET coordinating centre at the Health Protection Agency Centre for Infections in London. There, the details are entered into a central database, which is then searched for other cases that stayed at the same accommodation sites as those visited by the new case. Either a single or a cluster notification will be faxed to collaborators, and the appropriate database, which is then searched for other cases that stayed at the same microbiological diagnoses and travel history, and then report them to the site that has not been associated with any other cases of legionnaires' disease but more than two years previously [2].

**Figure 1**

EWGLI collaborating countries, 2004

Note: Where more than one collaborating centre is located in a town, only one point is shown.

**Table 1**

Countries reporting more than 10 cases of travel-associated legionnaires' disease in 2004, EWGLI

<table>
<thead>
<tr>
<th>Country of report</th>
<th>Number of cases</th>
</tr>
</thead>
<tbody>
<tr>
<td>England &amp; Wales</td>
<td>172</td>
</tr>
<tr>
<td>France</td>
<td>135</td>
</tr>
<tr>
<td>The Netherlands</td>
<td>119</td>
</tr>
<tr>
<td>Italy</td>
<td>66</td>
</tr>
<tr>
<td>Denmark</td>
<td>33</td>
</tr>
<tr>
<td>Spain</td>
<td>22</td>
</tr>
<tr>
<td>Sweden</td>
<td>22</td>
</tr>
<tr>
<td>Scotland</td>
<td>17</td>
</tr>
<tr>
<td>Austria</td>
<td>16</td>
</tr>
<tr>
<td>Belgium</td>
<td>12</td>
</tr>
</tbody>
</table>

Note: In addition, a number of countries reported fewer than 10 cases, and are not listed here.

The cases reported in 2004 generally fit the distinctive age and gender profile seen in previous years, with male cases outnumbering female cases by 2.9 to 1. The median age for male cases was 57 years (age range 23-96) and for female cases was 60 years (age range 29-84).

The usual pattern of a seasonal peak in summer was repeated in 2004, though with a single peak in August, rather than the July and September peaks witnessed in 2002 and 2003.

**Deaths**

Thirty seven deaths were reported to EWGLINET in 2004, representing a case fatality rate of 5.6% (6% in 2003), and an additional 41.5% of cases reportedly recovered from their illness (38% in 2003). Together these categories (death and recovery) are considered to be the 'known' outcomes, as opposed an 'unknown' outcome (52.8% of cases in 2004); the known outcomes making up a larger proportion of cases in 2004 (47.2%) than in 2003 (44%) or 2002 (36.1%). This continues to reverse the trend seen between 1995 and 2002 of a falling rate of known outcomes versus unknowns.

Thirty of the deaths were in men (81%), and seven in women (19%). All of the individuals who died were between 41 and 83 years old. Twenty five of the deaths were associated with single cases (68%), 12 with cluster cases (32%).

**Microbiology**

The proportion of cases in which detection of legionella urinary antigen was the main method of diagnosis increased to 84.9% in 2004 (81.5% in 2003). Diagnoses where the main method of detection was serology continued their decline on previous years, falling to 8.7% in 2004 (10.0% in 2003); the diagnoses were composed of 3.7% by four-fold rise and 5.0% by single high titre. The number of culture proven cases dropped to 37 (48 in 2003), representing just 5.6% of all cases. Five cases (0.8%) were diagnosed primarily by other methods.

Of the 37 deaths in 2004, seven were diagnosed primarily by culture (19%), 27 primarily by urinary antigen (73%), two by serology (four-fold rise) (5%), and one by direct immunofluorescence (3%). Twenty two of the deaths were caused by 'L. pneumophila serogroup 1' infection (69.4%), one was due to 'L. pneumophila other serogroup' (2%), nine were attributed to 'L. pneumophila serogroup unknown', four to 'Legionella unknown' (11%), and one to 'Legionella other species' (3%) (the species was not specified).

The main category of organism detected in 2004 was 'L. pneumophila serogroup 1' (454 cases, 69.3%). The remaining cases were reported...
as ‘L. pneumophila other serogroup’ (13 cases, 2.0%), ‘L. pneumophila serogroup unknown’ (154 cases, 23.5%), ‘Legionella other species’ (2 cases, 0.3%), and ‘Legionella species unknown’ (32 cases, 4.9%).

**Travel**

Although cases in 2004 visited around 60 different countries, over half (53%) were associated with travel to the four main countries of infection: France (126 cases), Italy (111), Spain (63), and Turkey (48) [FIGURE 2]. A large proportion of the cases visiting sites in France were French nationals, travelling internally in their own country, and likewise with Italian nationals visiting sites in Italy (54 cases). For cases involving travel in Spain, the proportion associated with clusters was 19%; for cases involving travel to France and Italy the figure was 23% for each, while for Turkey it was 44% (although this proportion is higher than that seen in the other three countries, it further consolidates the improvements seen on the 71% of cases in Turkey which were associated with clusters in 2002).

**Cluster**

Eighty six new clusters were identified in 2004, compared with 89 in 2003 and 94 in 2002 (this does not include clusters which were identified in previous years and were associated with a subsequent case in 2004; these clusters are included in the previous years’ figures). The size of these clusters varied less than in previous years, with the largest cluster involving six cases (down from 17 cases in 2003), although, as in previous years, the majority of clusters (59 in 2004) involved just two cases. There was a slight shift towards clusters involving three cases (up from nine in 2003 to 18 in 2004), but in 2004 the proportion of clusters involving only two or three cases reached almost 90%, compared with 84% in 2003 and 81% in 2002 [FIGURE 3]. Of the 86 clusters, 39 consisted of a single case reported by each of two or more countries. National surveillances schemes do not normally detect clusters that involve fewer than two of their citizens, and therefore would not ordinarily have detected these clusters.

In 2004, clusters were located in 24 countries, and one cluster was associated with a cruise ship [TABLE 2]. Italy and France were associated with the most clusters (17 clusters each, plus another cluster involving sites in both Italy and Germany), followed by Spain and Turkey which were each associated with nine clusters. Of the remaining clusters, the number occurring in countries outside EWGLINET, or in EWGLINET countries not officially signed up to follow the European guidelines, was 14 (representing 16%, an increase on the 13% seen in 2003, and following the trend of increased cluster detection outside the area of operation of the European guidelines). Five clusters involved two or more accommodation sites, including the one mentioned above which spanned two countries (Italy and Germany).

Most of the clusters in 2004 occurred during the summer months (66 between May and September, representing 77% of the full year figure). January was the only month in 2004 during which no clusters were detected.

**Figure 2**

Countries visited by more than 10 cases of travel-associated legionnaires’ disease in 2004, by case type, EWGLI 2004

- France
- Italy
- Spain
- Turkey
- England
- Greece
- Portugal
- Malta
- Germany
- USA
- Netherlands

Fifty five cases visited more than one European country, and ten cases visited more than one country outside Europe. An additional 66 cases (10.1%) visited countries outside the EWGLINET scheme.

**Clusters**

Eighty six new clusters were identified in 2004, compared with 89 in 2003 and 94 in 2002 (this does not include clusters which were identified in previous years and were associated with a subsequent case in 2004; these clusters are included in the previous years’ figures). The size of these clusters varied less than in previous years, with the largest cluster involving six cases (down from 17 cases in 2003), although, as in previous years, the majority of clusters (59 in 2004) involved just two cases. There was a slight shift towards clusters involving three cases (up from nine in 2003 to 18 in 2004), but in 2004 the proportion of clusters involving only two or three cases reached almost 90%, compared with 84% in 2003 and 81% in 2002 [FIGURE 3]. Of the 86 clusters, 39 consisted of a single case reported by each of two or more countries. National surveillances schemes do not normally detect clusters that involve fewer than two of their citizens, and therefore would not ordinarily have detected these clusters.

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**Figure 3**

Number of cases of travel-associated legionnaires’ disease per cluster, by year, EWGLI 2004

**Table 2**

Countries associated with clusters of travel-associated legionnaires’ disease in 2004, EWGLI

<table>
<thead>
<tr>
<th>Country of Infection</th>
<th>Number of clusters</th>
</tr>
</thead>
<tbody>
<tr>
<td>Austria</td>
<td>2</td>
</tr>
<tr>
<td>Bulgaria</td>
<td>1</td>
</tr>
<tr>
<td>Channel Islands</td>
<td>1</td>
</tr>
<tr>
<td>Cruise</td>
<td>1</td>
</tr>
<tr>
<td>Cuba</td>
<td>2</td>
</tr>
<tr>
<td>Dominican Republic</td>
<td>1</td>
</tr>
<tr>
<td>France</td>
<td>17</td>
</tr>
<tr>
<td>Germany</td>
<td>1</td>
</tr>
<tr>
<td>Greece</td>
<td>2</td>
</tr>
<tr>
<td>Hungary</td>
<td>1</td>
</tr>
<tr>
<td>Italy</td>
<td>17</td>
</tr>
<tr>
<td>Italy/Germany</td>
<td>1</td>
</tr>
<tr>
<td>Jordan</td>
<td>1</td>
</tr>
<tr>
<td>Malta</td>
<td>4</td>
</tr>
<tr>
<td>Mexico</td>
<td>1</td>
</tr>
<tr>
<td>The Netherlands</td>
<td>1</td>
</tr>
<tr>
<td>Poland</td>
<td>1</td>
</tr>
<tr>
<td>Portugal</td>
<td>4</td>
</tr>
<tr>
<td>Russia</td>
<td>1</td>
</tr>
<tr>
<td>Spain</td>
<td>9</td>
</tr>
<tr>
<td>Sri Lanka</td>
<td>2</td>
</tr>
<tr>
<td>Tunisia</td>
<td>3</td>
</tr>
<tr>
<td>Turkey</td>
<td>9</td>
</tr>
<tr>
<td>UAE</td>
<td>1</td>
</tr>
<tr>
<td>USA</td>
<td>1</td>
</tr>
<tr>
<td>Uzbekistan</td>
<td>1</td>
</tr>
</tbody>
</table>
Investigations and publications
A total of 96 sites were involved in the 86 new clusters in 2004. Of these sites, 17 were in countries not signed up to follow the European guidelines, and one site was already under investigation, leaving 78 that required EWGLINET investigations. Additionally, 15 sites that had been involved in clusters in previous years were associated with extra cases during 2004 (‘cluster updates’) and so needed to be re-investigated (one twice, resulting in a need for 16 re-investigations). These sites had been previously investigated under the guidelines, and are known as ‘re-offending’ sites.

In total, EWGLINET requested the investigation of 94 sites for clusters and cluster updates in 2004. Fifty three ‘Form B’ reports (56.4%) advised that samples from the accommodation site had tested positive for *L. pneumophila* (at concentrations equal to or greater than 1000 cfu/litre [5]), 38 (40.4%) reported that *L. pneumophila* was not detected in samples, and three ‘Form B’ reports (3.2%) did not have samples taken for reasons accepted by the coordinating centre.

The names of three French sites and one site in Turkey were published on the EWGLI website during 2004 for failure to return reports on time, or for failure to implement appropriate control measures in time. This represents a significant reduction from the 27 site names published during 2003.

During 2004, investigation reports were received for 149 sites associated with just a single case, even though the EWGLI guidelines do not require these. Of the 145 sites at which sampling was undertaken, 76 (52.4%) were reported positive for *L. pneumophila*.

Discussion
The EWGLINET surveillance scheme for travel-associated legionnaires’ disease has now been in operation for 17 years. Each year the scheme detects a large number of clusters that involved no more than one case from any country and would otherwise have gone undetected. Thirty nine such clusters were identified by EWGLINET in 2004 (45%), and were therefore subjected to the high standard of investigation and control demanded by the EWGLI guidelines.

Italy and France continue to report a high proportion of their internal travel cases (for example, cases in French people travelling within France). These cases are important because they allow EWGLINET to detect additional clusters within Italy and France that might otherwise go undetected. EWGLINET encourages other countries to do the same by ensuring that their internal travel cases are reported.

The number of postings on the EWGLI website dropped dramatically in 2004, demonstrating that countries (especially Turkey, who had a much higher number of sites published in 2003 than in 2004) have adapted well to implementing the guidelines in a timely fashion. It is especially promising to note that the proportion of smaller clusters (clusters involving just two or three cases) has increased since the introduction of the EWGLI guidelines, which suggests that the standard of investigation and control outlined in the guidelines has proven sufficient to prevent a large number of further cases developing from those accommodation sites.

There continue to be areas where surveillance could be improved across Europe. Data on deaths is not as detailed as it could be. Cases are often reported to EWGLINET as ‘still ill’ or ‘unknown’, and these cases may eventually be fatal. Unfortunately, EWGLINET is rarely updated on the status of these cases, and after a year they become classified as ‘outcome unknown’. Collaborators are encouraged to let the coordinating centre know the outcome of cases that were reported while the patient was still ill. The proportion of cases reported to the scheme with known outcomes has been increasing, which is promising.

Cultures were taken for 19% of fatalities, which is an improvement on the cultures taken in only 5.6% of cases overall, but this percentage is still lower than would be liked. Fatal cases are often investigated more thoroughly than cases in patients who recover, and in order to demonstrate that the infection came from a particular source, a clinical culture is required for each case. Clinicians should be encouraged to take samples for culture wherever possible, and especially in fatal cases.

The seasonal pattern typically seen by EWGLI each year, with a concentration of cases during the summer months, can be explained for the most part by the fact that the scheme records only travel associated cases of legionnaires’ disease, and the majority of people in Europe choose to take their holidays during the northern hemisphere summer. However, national surveillance systems, which deal with community and hospital-acquired cases as well as travel-associated cases, often see a marked increase in case numbers over the summer months that cannot be attributed solely to travel patterns. It may be that the warmer ambient temperatures in summer provide a more amenable environment for the legionella bacteria to multiply.

The surveillance scheme continues to expand to cover a greater number of European countries. The addition of Andorra to the scheme in 2004 brought the number of collaborating countries up to 37, but there are areas of eastern Europe that do not yet participate. It should be a priority for the scheme to form a working relationship with these countries with the intent of forming official collaborations with them at the earliest possible date, so that cases of travel-associated legionnaires’ disease occurring in their residents can be added to the European dataset.

Acknowledgements
This work is funded by the European Commission Health and Consumer Protection Directorate-General.

We would like to thank all the collaborators* for reporting their cases and all the people involved in public health control and prevention programs for travel-associated legionnaires’ disease.

* The list of EWGLI collaborators is available at the following URL address: www.ewgli.org/contact/contact_listof_collaborators.asp

References
The 2004-2005 influenza season in Europe started in late December 2004 and the first influenza-activity occurred in the west and southwest (Spain, United Kingdom and Ireland). Influenza activity then moved gradually east across Europe during January and early February 2005, and from late February until late March, most movement was south to north. The intensity of clinical-influenza activity in ten out of 23 countries was higher than during the 2003-2004 season, and lower or equal to the 2003-2004 season in the other 13 countries. The highest consultation rates were generally observed among children aged 0-14 years. However, the peak consultation rates due to influenza-like illness or acute respiratory infection were not especially high when compared with historical data. The predominant virus strain was influenza A (83% of total detections) of the H3 subtype (85% of H-subtyped A viruses), with fewer influenza B (17% of total detections) or A(H1) viruses (15% of H-subtyped A viruses) detected. The vast majority of A(H3) viruses were similar to the reference strains A/Wellington/1/2004 (H3N2) and, subsequently, A/California/7/2004 (H3N2) that are closely related drift variants of the A/Fujian/411/2002 (H3N2) prototype vaccine strain. The B viruses co-circulated with A viruses during the whole influenza season in 11 out of 24 countries. Seven of these were located in the northeast of Europe and in these general practices the proportion of B viruses was higher (range: 31-60%) than in the rest of Europe (range: 6-26%). In 13 out of 24 countries the B viruses circulated relatively late in the season. About 43% of all antigenically characterised B viruses were B/Hong Kong/330/2001-like (B/Victoria/2/87 lineage), a strain that is distinguishable from the vaccine influenza B strain, which was a B/Yamagata/16/88 lineage virus. Based on the viruses detected worldwide until February 2005, the World Health Organization modified the composition of the 2005-2006 influenza vaccine from the 2004-2005 season vaccine to include a new A(H3N2) component: an A/California/7/2004 (H3N2)-like virus.

**Keywords:** Epidemiology, Europe, Influenza, Surveillance, Virology

**Introduction**

Influenza has a considerable public health impact in Europe each winter. Seasonal epidemics are associated with higher general practice consultation rates [1], increased hospital admissions [2] and excess deaths [2, 3].

The European Influenza Surveillance Scheme (EISS) is a collaborative project of physicians (mainly in primary care), epidemiologists and virologists, and aims to contribute to a reduction in morbidity and mortality due to influenza in Europe by active clinical and virological surveillance of influenza [4-6]. The participating national reference laboratories have functioned within EISS as the Community Network of Reference Laboratories for Human Influenza in Europe (CNRL) since 2003 [7]. An important objective for the scheme has been the inclusion of all members of the European Union (EU), as required by EU Decision 2119/98/EC on the establishment of dedicated surveillance networks for communicable diseases [8], and this was achieved at the end of the 2004-2005 season.

Including all members who participated in EISS during the 2004-2005 season (20 EU countries, Norway, Romania and Switzerland), the EISS project comprised 30 national influenza reference laboratories. The characteristics of the sentinel networks during the 2004-2005 season are summarised in Table 1. The median weekly population under clinical surveillance by the sentinel networks during the 2004-2005 season varied from 0.4% to 100% of the total population of a country, representing at least a median number of 17.8 million inhabitants of Europe [TABLE 1]. The sentinel surveillance is carried out by 12 902 general practitioners (GPs), paediatricians and other physicians, although during the 2004-2005 season the number of physicians reporting each week was often lower than this [TABLE 1]. In general, the age distribution of the population under surveillance is representative for the age distribution of the total population in a country, although in some countries the population under surveillance is skewed to the lower ages (partly due to a high proportion of paediatricians) and/or higher ages [TABLE 1]. Further data about representativeness of the population under surveillance in EISS can be found for most countries in Aguilera et al. [11].

A proportion of the sentinel physicians, in general representative for the surveillance network in a country, also collects nose and/or throat swabs for virological surveillance using a swabbing protocol that guarantees representative swabbing during the season [TABLE 1] [11]. Combining clinical and virological data in the same population allows the validation of clinical reports made by the sentinel physicians and provides virological data in a clearly defined population, the general population that visits a physician with an influenza-like illness (ILI) or acute respiratory infection (ARI) [12]. In addition to specimens obtained from physicians in the sentinel surveillance systems, the laboratories also collect and report results on specimens obtained from other sources (e.g. from hospitals or non-sentinel physicians). These data are called ‘non-sentinel’ in this paper and are collected to give a second measure of influenza activity and to analyse the representativeness of the virological data obtained from the sentinel physicians [12]. Based on the collection of virological data, the total population under surveillance of EISS was about 462 million inhabitants of Europe during the 2004-2005 season.

The identification of circulating viruses within the population and the recognition of virological changes are important tasks for EISS in order to fulfil its early warning function [7]. There is a particular need to detect and monitor the emergence or re-emergence of viruses with pandemic potential and viruses that have a ‘mismatch’ with the vaccine strain components, and to monitor their clinical impact.

This report presents an analysis and interpretation of influenza surveillance data collected by European countries that were active members of EISS during the 2004-2005 season.

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1. European Influenza Surveillance Scheme Co-ordination Centre, Netherlands Institute for Health Services Research (NIVEL), Utrecht, the Netherlands
2. Current affiliation: WHO Regional Office for Europe, Copenhagen, Denmark
3. Radboud University Medical Center, Nijmegen, The Netherlands
4. EISS members (2004-2005 season)
Table 1: Some characteristics of the national sentinel surveillance networks during the 2004-2005 season

<table>
<thead>
<tr>
<th>Country</th>
<th>GPs</th>
<th>Paediatricians</th>
<th>Other a</th>
<th>No. of physicians in the sentinel networks</th>
<th>No. of physicians that reported ILI/ARI during the season</th>
<th>Population under surveillance during the season</th>
<th>% of total population b</th>
<th>Age distribution; median % c,d,e</th>
<th>Age distribution total; population; % f,g,h</th>
<th>% of sentinel physicians who took swabs i</th>
</tr>
</thead>
<tbody>
<tr>
<td>Austria</td>
<td>42</td>
<td>14</td>
<td>–</td>
<td>38</td>
<td>18-47</td>
<td>Median</td>
<td>0.7</td>
<td>0.3-0.9</td>
<td>16</td>
<td>68</td>
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<td>Belgium</td>
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<td>–</td>
<td>–</td>
<td>39</td>
<td>29-44</td>
<td>0.4</td>
<td>0.3-0.5</td>
<td>18</td>
<td>66</td>
<td>17</td>
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<td>2230</td>
<td>1240</td>
<td>–</td>
<td>3115</td>
<td>3036-3181</td>
<td>47.3</td>
<td>46.2-48.3</td>
<td>18</td>
<td>64</td>
<td>18</td>
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<td>150</td>
<td>–</td>
<td>–</td>
<td>125</td>
<td>98-143</td>
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<td>2.7-4.0</td>
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<td>66</td>
<td>15</td>
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<td>360</td>
<td>–</td>
<td>–</td>
<td>294</td>
<td>152-319</td>
<td>1.1</td>
<td>0.5-1.2</td>
<td>18</td>
<td>67</td>
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<td>378</td>
<td>7</td>
<td>–</td>
<td>376</td>
<td>282-415</td>
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<td>0.6-0.7</td>
<td>23</td>
<td>61</td>
<td>16</td>
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<td>Germany</td>
<td>604</td>
<td>146</td>
<td>33</td>
<td>593</td>
<td>437-639</td>
<td>1.6</td>
<td>1.2-1.7</td>
<td>22</td>
<td>55</td>
<td>23</td>
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<td>Ireland</td>
<td>68</td>
<td>–</td>
<td>–</td>
<td>61</td>
<td>52-68</td>
<td>2.5</td>
<td>2.2-2.7</td>
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<td>n.k.</td>
<td>n.k.</td>
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<td>750</td>
<td>100</td>
<td>–</td>
<td>399</td>
<td>238-859</td>
<td>0.9</td>
<td>0.5-2.1</td>
<td>18</td>
<td>63</td>
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<td>113</td>
<td>–</td>
<td>–</td>
<td>n.k.</td>
<td>n.k.</td>
<td>8.7</td>
<td>n.a</td>
<td>19</td>
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<td>16</td>
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<td>Lithuania</td>
<td>321</td>
<td>327</td>
<td>396</td>
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<td>39.7</td>
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<td>4</td>
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<td>13</td>
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<td>0.9</td>
<td>0.4-1.2</td>
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<td>n.k.</td>
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<td>Malta</td>
<td>22</td>
<td>–</td>
<td>–</td>
<td>22</td>
<td>n.k.</td>
<td>n.k.</td>
<td>n.k.</td>
<td>n.k.</td>
<td>n.k.</td>
<td>18</td>
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<tr>
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<td>67</td>
<td>–</td>
<td>–</td>
<td>41</td>
<td>37-44</td>
<td>0.6</td>
<td>0.4-0.9</td>
<td>18</td>
<td>69</td>
<td>14</td>
</tr>
<tr>
<td>Northern Ireland</td>
<td>93</td>
<td>–</td>
<td>–</td>
<td>75</td>
<td>60-88</td>
<td>7.0</td>
<td>5.7-7.8</td>
<td>20</td>
<td>66</td>
<td>14</td>
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<tr>
<td>Norway</td>
<td>–</td>
<td>201</td>
<td>n.k.</td>
<td>n.k.</td>
<td>n.k.</td>
<td>n.k.</td>
<td>n.k.</td>
<td>14</td>
<td>63</td>
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<td>–</td>
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<td>144-219</td>
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<td>18</td>
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<td>Portugal</td>
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<td>–</td>
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<td>20-68</td>
<td>0.6</td>
<td>0.3-1.0</td>
<td>16</td>
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<td>19</td>
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<td>102</td>
<td>–</td>
<td>225</td>
<td>206-240</td>
<td>2.2</td>
<td>1.7-2.2</td>
<td>28</td>
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<td>75</td>
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<td>7.0</td>
<td>5.7-7.8</td>
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<td>14</td>
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<td>Slovakia</td>
<td>2121</td>
<td>1202</td>
<td>–</td>
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<td>100</td>
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<td>19</td>
<td>65</td>
<td>16</td>
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<td>Slovenia</td>
<td>14</td>
<td>12</td>
<td>12</td>
<td>36</td>
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<td>391</td>
<td>102</td>
<td>–</td>
<td>n.k.</td>
<td>n.k.</td>
<td>1.3</td>
<td>0.6-1.4</td>
<td>18</td>
<td>63</td>
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<td>Sweden</td>
<td>–</td>
<td>96</td>
<td>64</td>
<td>36-72</td>
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<td>n.k.</td>
<td>n.k.</td>
<td>n.k.</td>
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<td>Switzerland</td>
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<td>43</td>
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<td>194</td>
<td>165-220</td>
<td>3.0</td>
<td>2.4-5.4</td>
<td>21</td>
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<td>15</td>
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<tr>
<td>Wales</td>
<td>30</td>
<td>–</td>
<td>–</td>
<td>n.k.</td>
<td>n.k.</td>
<td>7.4</td>
<td>7.2-7.4</td>
<td>17</td>
<td>64</td>
<td>19</td>
</tr>
</tbody>
</table>

1. Number of physicians reporting ILI/ARI and population under surveillance are based on weekly reports of these figures during the 2004-2005 season. ILI = Influenza-Like Illness; ARI = Acute Respiratory Infection; GPs = general practitioners; n.k. = not known; n.a. = not applicable.
2. Total population figures and age distribution were derived from reference [1] for all countries except the United Kingdom. Data for all countries except Belgium and Italy were from 1 January 2005, for Belgium and Italy from 1 January 2004. For the United Kingdom, data for England, Northern Ireland, Scotland and Wales was used; total population figures are from 2004 and the age distribution is from the Census 2001.
3. Malta and Norway record encounters. The age distribution for Malta was calculated from the proportion of the population under surveillance for which the age was known.
4. Totals may not sum to 100 due to rounding.
6. Germany and Switzerland: Internists; Slovenia: "community practitioners" for 7 to 18 years-old; Lithuania: therapists; Norway and Sweden: practitioners.
7. No or partial overlap with physicians/practitioners collecting clinical data.
8. 67% of physicians agreed to take swabs, however, due to the mild season 38% of physicians actually took swabs during the 2004-2005 season.
9. All GPs and paediatricians in Slovakia are obliged to report virus isolates during the 2004-2005 season.
10. Sentinel physicians also obtained nasal, pharyngeal, or nasopharyngeal specimens from a subset of patients and these were sent to the national reference laboratory or laboratories for virological analysis. The laboratories also collected and reported results on specimens obtained from other sources (e.g. from hospitals or non-sentinel physicians).
11. The virological data included results mostly from cell cultures followed by virus type and subtype identification and from rapid diagnostic enzyme-immunological or immunofluorescence tests identifying the virus type only. Many laboratories also routinely use reverse transcription polymerase chain reaction (RT-PCR) for detection, typing and subtyping [13]. About 75% (20/26) of the countries reported antigenic characterisation data and almost 50% (12/26) of the countries reported genetic characterisation data of the virus isolates during the 2004-2005 season.

Methods

Twenty-six countries actively monitored influenza activity from week 40/2004 (27/9/2004- 3/10/2004) to week 20/2005 (16/5/2005 - 22/5/2005) during the 2004-2005 season [TABLE 1] (in this paper England, Northern Ireland, Scotland and Wales were considered as four separate countries as they each have their own surveillance system). This paper only presents data collected until week 16/2005 (12/26) of the countries reported genetic characterisation data of the virus isolates during the 2004-2005 season.
During the influenza season, the weekly clinical and virological data were processed and analysed by the national centres and then entered into the EISS database the following week via the internet (www.eiss.org) [14]. The indicators of influenza activity were established on a weekly basis by the national coordinators: the intensity of clinical activity and the geographical spread of influenza (see Box), and the dominant type/subtype circulating in the population (definition not shown). The dominant type/subtype for the season as a whole was estimated per country using the algorithm shown in the box. During the 2004-2005 season eight countries entered a baseline (see Box).

**Box. Definitions of indicators**

**Baseline**
Level of clinical influenza activity calculated nationally representing the level of clinical activity in the period that the virus is not epidemic (summer and most of the winter) based on historical data (5-10 influenza seasons).

**Intensity**
The intensity of clinical activity compares the weekly clinical morbidity rate with historical data:

- Low – no influenza activity or influenza activity at baseline level
- Medium – usual levels of influenza activity
- High – higher than usual levels of influenza activity
- Very high – particularly severe levels of influenza activity (less than once every 10 years)

**Geographic spread**
The geographic spread is a WHO indicator that has the following levels:

- No activity – no evidence of influenza virus activity (clinical activity remains at baseline levels)
- Sporadic – isolated cases of laboratory confirmed influenza infection
- Local outbreak – increased influenza activity in local areas (e.g. a city) within a region, or outbreaks in two or more institutions (e.g. schools) within a region; laboratory confirmed
- Regional activity – influenza activity above baseline levels in one or more regions with a population comprising less than 50% of the country’s total population; laboratory confirmed,
- Widespread – influenza activity above baseline levels in one or more regions with a population comprising 50% or more of the country’s population, laboratory confirmed

**Dominant virus**
The assessment of the dominant virus for the season is based on:

- Sentinel and non-sentinel data (primary assessment sentinel data)
- A minimum number of 10 isolates
- If more than 10% of total A isolates are H-subtyped the H subtype is taken into consideration
- If more than 10% of total A isolates are N-subtyped the N subtype is also taken into consideration
- The limits for co-dominant virus types/subtypes are: 45%-55%

**Results**
The 2004-2005 influenza season in Europe began in December 2004 and clinical influenza activity first occurred in the southwest (United Kingdom, Spain and Ireland) and gradually moved east across Europe, starting in Italy/Portugal, France/Switzerland, Austria/Luxembourg, Slovenia/Czech Republic/the Netherlands/Belgium/Germany in subsequent weeks during January 2005 (see Figure 1 at http://www.eiss.org/documents/eurosurveillance_supplement_2004-2005_season.pdf). Thereafter, influenza activity moved in a more southerly-northerly direction starting in Poland/Lithuania/Sweden, Denmark/Norway and Romania/Slovakia/Latvia in subsequent weeks from February until March. A similar movement was seen when the timing of peak clinical influenza activity across Europe was analysed. By regression analysis of plots of the longitude and latitude of the centre of each country against the week of peak influenza activity, both the west-east (R2 = 0.6796; p<0.001) and south-north (R2 = 0.2496; p=0.018) movement were statistically significant. The timing is nicely visualised in figure 1.

**Figure 1**
Timing of peak clinical influenza activity across Europe during the 2004-2005 season

The peak intensity of clinical influenza activity ranged from low in Scotland and Wales to high in ten countries, and 15 of 25 countries reported widespread influenza activity during the 2004-2005 season [TABLE 2] (see also Figure 1 at http://www.eiss.org/documents/eurosurveillance_supplement_2004-2005_season.pdf). The peak levels of ILI/ARI consultation rates in Europe were reached between week 50/2004 and 12/2005 [TABLE 2], covering a period of 13 weeks between the first and last peak. The week of peak ILI/ARI consultation rates coincided roughly with the peak of sentinel influenza virus detections [TABLE 2]. A detailed breakdown of the sentinel clinical and virological data by week and country is available from the EISS website (see Figure 2 at http://www.eiss.org/documents/eurosurveillance_supplement_2004-2005_season.pdf).

In countries reporting age specific data (N=20), the highest consultation rates during the influenza peak were observed among children in the age groups 0-4 years and 5-14 years in 12 countries [TABLE 2]. In four of these countries the consultation rate was slightly higher in the 5-14 age group than in the 0-4 age group.
and in the other eight countries the consultation rate was slightly higher in the 0-4 age group than in the 5-14 age group [TABLE 2]. In Austria and Northern Ireland the consultation rate was clearly highest in the 0-4 age group. Although in the Netherlands, Norway, Portugal and Romania the consultation rate was also high in the younger age groups, in the Netherlands and Portugal the consultation rate was highest among people aged 65+ years in one week and in Norway and Romania the consultation rate was also high in the 15-64 years age group [TABLE 2].

For Europe as a whole, the largest number of positive specimens was detected between week 5/2005 and 11/2005 [FIGURE 2]. A total of 15,295 sentinel and non-sentinel specimens were positive for influenza virus: 12,745 (83%) were influenza A and 2,550 (17%) were influenza B. Of all haemagglutinin-subtyped viruses (N=6,648), 5,651 (85%) were H3 and 997 (15%) were H1. All 2,102 neuraminidase-subtyped A(H3) viruses were of the N2 subtype and of the 467 neuraminidase-subtyped A(H1) viruses 465 (99%) were N1 and only about 1% (2 viruses) N2. The predominant virus circulating in the individual countries was mostly influenza A(H3) [TABLE 2]. The B viruses co-circulated the whole season with A viruses in 11 out of 24 countries [TABLE 2]. Seven of these countries were located in the northeast of Europe and the proportion of B viruses in this region was higher (range: 31%-60%) than in the rest of Europe (range: 6%-26%) [TABLE 3]. In 13 out of 24 countries, the B viruses circulated relatively late in the season [TABLE 3]. The distribution of B viruses over sentinel and non-sentinel sources was variable [TABLE 3]. A detailed breakdown by country of the virological data collected in the sentinel and non-sentinel systems is available from the EISS website (see Figure 2.

![Table 2](https://example.com/table2.png)

**Table 2**

Overview of influenza activity during the 2004-2005 season

<table>
<thead>
<tr>
<th>Country (N=26)</th>
<th>Week(s) of peak clinical activity</th>
<th>Most affected age groups</th>
<th>Intensity (peak level)</th>
<th>Week(s) of peak virus detections</th>
<th>Dominant virus type/subtype</th>
<th>Geographical spread (peak level)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Influenza-like illness:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>England</td>
<td>No peak</td>
<td>None</td>
<td>Medium</td>
<td>5</td>
<td>A(H3N2)</td>
<td>Regional</td>
</tr>
<tr>
<td>Scotland</td>
<td>No peak</td>
<td>n.a.</td>
<td>Low</td>
<td>5 + 10</td>
<td>A(H3)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Wales</td>
<td>No peak</td>
<td>None</td>
<td>Low</td>
<td>7</td>
<td>A</td>
<td>Widespread</td>
</tr>
<tr>
<td>Northern Ireland</td>
<td>50 + 1</td>
<td>0-4</td>
<td>Medium</td>
<td>n.a.</td>
<td>A(H3)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Ireland</td>
<td>1</td>
<td>n.a.</td>
<td>Medium</td>
<td>53</td>
<td>A(H3N2)</td>
<td>Local</td>
</tr>
<tr>
<td>Spain</td>
<td>2-3</td>
<td>5-14, 0-4</td>
<td>High</td>
<td>2</td>
<td>A(H3)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Portugal</td>
<td>5</td>
<td>5-14, 65+</td>
<td>High</td>
<td>4</td>
<td>A(H3)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Belgium</td>
<td>6-8</td>
<td>5-14, 0-4</td>
<td>Medium</td>
<td>9</td>
<td>A(H3N2)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Italy</td>
<td>6</td>
<td>0-4, 5-24</td>
<td>High</td>
<td>5</td>
<td>A(H3N2)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Austria</td>
<td>7</td>
<td>0-4</td>
<td>High</td>
<td>9</td>
<td>A(H3N2)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Luxembourg</td>
<td>7</td>
<td>n.a.</td>
<td>High</td>
<td>7</td>
<td>A(H3N2)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Netherlands</td>
<td>7</td>
<td>0-4, 65+</td>
<td>High</td>
<td>7</td>
<td>A(H3)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Slovenia</td>
<td>7</td>
<td>0-4, 5-24</td>
<td>Medium</td>
<td>8</td>
<td>A(H3N2) + B</td>
<td>Widespread</td>
</tr>
<tr>
<td>Malta</td>
<td>8-9</td>
<td>n.a.</td>
<td>n.a.</td>
<td>n.a.</td>
<td>n.a.</td>
<td>n.a.</td>
</tr>
<tr>
<td>Poland</td>
<td>8-11</td>
<td>0-4, 5-24</td>
<td>High</td>
<td>10</td>
<td>A(H3) + B</td>
<td>Regional</td>
</tr>
<tr>
<td>Denmark</td>
<td>11</td>
<td>0-4, 5-24</td>
<td>High</td>
<td>8</td>
<td>A(H3N2)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Latvia</td>
<td>11-12</td>
<td>0-4, 5-24</td>
<td>Medium</td>
<td>9</td>
<td>A(H3)</td>
<td>Regional</td>
</tr>
<tr>
<td>Lithuania</td>
<td>11</td>
<td>0-4, 5-24</td>
<td>High</td>
<td>n.a.</td>
<td>n.a.</td>
<td>Regional</td>
</tr>
<tr>
<td>Romania</td>
<td>11</td>
<td>15-64, 5-14</td>
<td>Medium</td>
<td>11</td>
<td>A(H3N2)</td>
<td>Regional</td>
</tr>
<tr>
<td>Slovakia</td>
<td>11</td>
<td>5-14, 0-4</td>
<td>Medium</td>
<td>10</td>
<td>A(H3) + B</td>
<td>Local</td>
</tr>
<tr>
<td>Sweden</td>
<td>11</td>
<td>n.a.</td>
<td>Medium</td>
<td>9</td>
<td>A</td>
<td>Widespread</td>
</tr>
<tr>
<td>Norway</td>
<td>12</td>
<td>5-14, 15-64</td>
<td>Medium</td>
<td>7</td>
<td>A(H3N2)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Acute respiratory infections:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>France</td>
<td>6</td>
<td>0-4, 5-24</td>
<td>Medium</td>
<td>5</td>
<td>A(H3N2)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Germany</td>
<td>7-9</td>
<td>0-4, 5-24</td>
<td>High</td>
<td>10</td>
<td>A(H3)</td>
<td>Widespread</td>
</tr>
<tr>
<td>Czech Republic</td>
<td>8</td>
<td>0-4, 5-24</td>
<td>Medium</td>
<td>9</td>
<td>A</td>
<td>Widespread</td>
</tr>
</tbody>
</table>

1. Sentinel data, except for dominant virus type/subtype for which sentinel and non-sentinel data were taken into account. For definitions of indicators see the Box n.a. = not applicable as no data was available or insufficient data was available. No peak = activity was not above baseline or was flat during the whole season
2. If two age groups are shown the sequence is: most affected, second most affected
3. Estimated primarily taking into account the percentage of influenza virus positive specimens and secondarily the absolute number of isolates when the percentage of positive specimens was ambiguous

![Figure 2](https://example.com/figure2.png)

**Figure 2**

Total number of sentinel and non-sentinel specimens positive for influenza viruses by week for Europe as a whole during the 2004-2005 season

and in the other eight countries the consultation rate was slightly higher in the 0-4 age group than in the 5-14 age group [TABLE 2]. In Austria and Northern Ireland the consultation rate was clearly highest in the 0-4 age group. Although in the Netherlands, Norway, Portugal and Romania the consultation rate was also high in the younger age groups, in the Netherlands and Portugal the consultation rate was highest among people aged 65+ years in one week and in Norway and Romania the consultation rate was also high in the 15-64 years age group [TABLE 2].

For Europe as a whole, the largest number of positive specimens was detected between week 5/2005 and 11/2005 [FIGURE 2]. A total of 15,295 sentinel and non-sentinel specimens were positive for influenza virus: 12,745 (83%) were influenza A and 2,550 (17%) were influenza B. Of all haemagglutinin-subtyped viruses (N=6,648), 5,651 (85%) were H3 and 997 (15%) were H1. All 2,102 neuraminidase-subtyped A(H3) viruses were of the N2 subtype and of the 467 neuraminidase-subtyped A(H1) viruses 465 (99%) were N1 and only about 1% (2 viruses) N2. The predominant virus circulating in the individual countries was mostly influenza A(H3) [TABLE 2]. The B viruses co-circulated the whole season with A viruses in 11 out of 24 countries [TABLE 2]. Seven of these countries were located in the northeast of Europe and the proportion of B viruses in this region was higher (range: 31%-60%) than in the rest of Europe (range: 6%-26%) [TABLE 3]. In 13 out of 24 countries, the B viruses circulated relatively late in the season [TABLE 3]. The distribution of B viruses over sentinel and non-sentinel sources was variable [TABLE 3]. A detailed breakdown by country of the virological data collected in the sentinel and non-sentinel systems is available from the EISS website (see Figure 2.

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Twenty one of the 26 countries reported antigenic and/or genetic characterisation of the haemagglutinin for a total of 4,253 virus isolates. Of the 3,964 antigenically characterised isolates, 179 were also genetically characterised. An additional 289 isolates were characterised genetically only. In total (N=4,253), the haemagglutinin of 1,604 (38%) viruses was reported as A/Wellington/1/2004 (H3N2)-like, of 1,012 (24%) as A/California/7/2004 (H3N2)-like, 92 (2%) as A/Fujian/411/2002 (H3N2)-like, two (0.05%) as A/Panama/2007/99 (H3N2)-like, 774 (18%) as A/New Caledonia/20/99 (H1N1)-like, 437 (10%) as B/Jiangsu/10/2003-like (B/Yamagata/16/88 lineage) and 332 (8%) as B/Hong Kong/330/2001-like (B/Victoria/2/87 lineage).

In countries reporting influenza B characterisations, influenza B/Hong Kong/330/2001-like viruses were always reported in combination with B/Jiangsu/10/2003-like viruses [TABLE 3].

### Discussion

The 2004-2005 influenza season in Europe began in December 2004, which was late in comparison to the previous season, which began in October/November 2003 [6]. Peak clinical influenza activity was, for all countries with the exception of Italy and Germany, more than five weeks later than in the 2003-2004 season. The 2004-2005 season was dominated by the spread of a drift variant relative to the A/Fujian/411/2002 (H3N2)-like virus that circulated in the 2003-2004 season, represented by the reference strains A/Wyoming/3/2003. Ninety-two viruses (2%) had an H3 antigenically similar to A/Fujian/411/2002 (H3N2). Two viruses had an H3 antigenically similar to the former vaccine strain A/Panama/2007/99 (H3N2). The H1 of 759 (19%) viruses was antigenically similar to the 2004-2005 vaccine strain A/New Caledonia/20/99 (H1N1). Among the 759 antigenically characterised B viruses, 433 (57%) were B/Jiangsu/10/2003-like and 326 (43%) were B/Hong Kong/330/2001-like.
The general progress of influenza activity across Europe during the 2004-2005 season differed from most previous seasons in that there was a west-east movement at the beginning of the season changing into a south-north movement later on in the season. Analysis of five previous seasons (1999-2000 to 2003-2004) indicated that there was a west-east movement of influenza activity in three seasons (2001-2002, 2002-2003 and 2003-2004), but that in the 2001-2002 season there was also a south-north movement similar to that found for the 2004-2005 season [18]. These analyses were done by plotting the longitude and latitude of the centre of each country against the week of peak incidence. Recently, Saito et al [15] applied the method of kriging to influenza data and as presented in this paper [FIGURE 1] this method has the advantage of visual presentation of the timing of peak clinical influenza activity on the map of Europe. The European map generated [FIGURE 1] indicates different timing in individual countries, which may be an artefact, as only the coordinates of the centre of a country were included. However, practice-based data from Germany indicated a similar south-north/east pattern as that observed in the EISS European analysis [19]. EISS is currently working on the extension of the method applied on the German data to include more European countries. In addition, further research is needed to determine what drives the direction of the movement or timing, such as type, subtype and antigenic characteristics of the founder virus, humidity, temperature, UV radiation and air traffic.

Although the age groups most affected were 0-4 years and 5-14 years, it should be noted that the estimated consultation rates for the different age groups are influenced by several factors such as consultation behaviour, estimation procedure, case definition, vaccination coverage and obligatory doctors visit for absence from work or school, which may differ between countries.

The continuous drift of the A(H3N2) viruses has led to the selection of the new reference viruses A/Wellington/1/2004 (H3N2) and A/California/7/2004 (H3N2), and both were reported to EISS during the 2004-2005 season. However, reference reagents for the antigenic characterisation of A/California/7/2004 (H3N2)-like viruses became available only halfway through the season, and retrospective analysis of a number of isolates from early in the season showed that a majority of these also resembled A/California/7/2004-like rather than A/Wellington/1/2004 (H3N2)-like. It is therefore possible that many of the viruses from the beginning of the season, which were recorded as A/Wellington/1/2004 (H3N2)-like at the time, actually belonged to the A/California/7/2004 (H3N2) drift variant. A recent analysis using antigenic cartography with data from the Netherlands and from the World Health Organisation (WHO) reference strains clearly showed the antigenic drift; when compared with large jumps of the A(H3N2) virus in the past, however, the recent drift was small and did not have a large clinical impact [20].

The influenza B virus detection results clearly demonstrated that there are differences between specimens collected from sentinel patients and non-sentinel patients. In only eight out of 19 countries was the proportion of B virus detections similar in sentinel and non-sentinel specimens. In eight other countries, most B virus detections were done in sentinel specimens, and in three countries, most detections were done in non-sentinel specimens [TABLE 3]. As influenza B virus infections are mostly mild and patients with these infections generally do not visit and are not admitted to hospitals, differences in the professions of doctors included in the sentinel and non-sentinel systems may explain these differences [21]. Another explanation might be the differences in age distribution of the population under surveillance in the sentinel systems [TABLE 1] and the differences in age distribution of the patients from whom a swab is taken. There are sentinel systems where a high proportion of specimens come from children, while others have a more balanced age distribution [21]. More systematic research into the structures of the various surveillance systems is needed to support these explanations.

Influenza B viruses currently circulating are antigenically and genetically divided into two distinct lineages represented by B/Yamagata/16/88 and B/Victoria/2/87 viruses, which have evolved to such an extent that antibodies raised to viruses of one lineage offer reduced cross-reactive protection against viruses of the other lineage [22,23]. The trivalent influenza vaccine, however, contains only one B virus component. Between 1990 and 2001, B/Yamagata/16/88 lineage viruses circulated worldwide and B/Victoria/2/87 lineage viruses circulated only in Asia. Since 2001, however, B/Victoria/2/87 lineage viruses have predominated in many countries, including in Europe, and the vaccine strain was changed accordingly. As B/Yamagata/16/88 lineage viruses predominated in the 2003-2004 season, a B/Yamagata/16/88 lineage virus was included in the northern hemisphere vaccine for the 2004-2005 season. In the 2004-2005 season there were more influenza B virus detections in Europe than in the 2003-2004 season: 15% compared with 0.9% [6]. In addition, 43% of the viruses belonged to the B/Victoria/2/87 lineage that was not included in the vaccine, and in five countries, the proportion of B/Victoria/2/87 lineage viruses among total B virus detections was higher than 50% (range 64-83%) [TABLE 3]. Notably, the 2005 season in New Zealand was dominated by circulation of influenza B viruses (almost 90% of total influenza viruses) and most of these belonged to the B/Victoria/2/87 lineage (almost 80% of the total number of characterised B viruses), which was also not included in the vaccine for the 2005 southern hemisphere season [24,25]. However, despite that, the clinical impact was less severe than that from the predominant circulation of A/Fujian/411/2002 (H3N2)-like viruses in the 2004 season in New Zealand [25,26]. In Australia, in contrast, mainly influenza A(H3) viruses (74% of all isolates) circulated during the 2005 season [24]. In the United States, about a quarter of all influenza viruses isolated during the 2004-2005 season were of the B type and, of the antigenically characterised B viruses, about 75% belonged to the B/Yamagata/16/88 lineage (strain in the vaccine) and 25% to the B/Victoria/2/87 lineage [27]. Since by February 2005 most B viruses isolated in the world were of the B/Yamagata/16/88 lineage type, the vaccine for the 2005-2006 northern hemisphere season again contains a B/Shanghai/361/2002-like virus (B/Yamagata/16/88 lineage) similar to the 2003-2004 season [22,28]. Since by September 2005 most B viruses belonged to the B/Victoria/2/87 lineage, the B/Victoria/2/87 lineage virus B/Malaysia/2506/2004 will be included in the vaccine for the 2006 southern hemisphere season [23]. Preliminary results show that B/Victoria/2/87 lineage viruses are predominating during the 2005-2006 season in Europe [29].

The WHO announced the composition of the influenza vaccine for the 2005-2006 northern hemisphere season in February 2005 [22]. Based on the analysis of influenza virus from all over the world up until February 2005, the A/Fujian/411/2002 (H3N2)-like vaccine strain in the influenza vaccine of 2004-2005 has been exchanged for a more recent virus: an A/California/7/2004 (H3N2)-like virus. In Europe, the vaccine composition recommended by the European Agency for the Evaluation of Medicinal Products, which is based on the WHO recommendations, has been used during the vaccine campaigns for the 2005-2006 season in Europe [28].

During the 2004-2005 season the A(H5N1) influenza virus causing epizootics in Asia and transmission to humans with fatalities [30] was not detected in poultry or humans in Europe. However, A(H5N1) infected birds smuggled into Belgium [31] and the by accidental worldwide distribution of an A(H2N2) virus in a quality control panel [32] in autumn 2004, highlighted the threat of introduction of a potential pandemic virus in Europe. Rapid inventories on the
level of laboratory preparedness carried out by the EISS coordination centre in January 2005 revealed that 26 of 32 national reference laboratories for human influenza and 22 of 25 European countries were prepared for detection of the A(H5N1) virus. However, only 12 of the laboratories were able to detect or identify specifically the A(H2) virus. The establishment of the CNRL and virology task groups strengthened the preparedness level of EISS as a whole by providing organised support through distribution of up to date RT-PCR detection protocols, recent sequence information, A(H5) controls for RT-PCR detection and the establishment of a reagent and sequence database [7]. These preparations proved useful when the A(H5N1) virus was recently introduced in many countries in Europe, probably by migrating birds, causing infections of wild birds and poultry [33], and since January 2006, human infection in Turkey [34].

The virological, epidemiological and clinical experts within EISS have been carefully monitoring the spread of virus strains in Europe during the 2005-2006 season. Assessment of the influenza activity is made in collaboration with the WHO Collaborating Centre in London and the European Centre for Disease Control and Prevention and is reported on the EISS website on a weekly basis.

Contributors

The members of EISS contributed by weekly submission of influenza surveillance data to EISS during the 2004-2005 season. CS Brown, TJ Meerhoff, A Meijer and WJ Paget carried out weekly analysis of the data and published the Weekly Electronic Bulletins during the 2004-2005 season. TJ Meerhoff extracted the clinical and virological data from the EISS databases for the paper and drafted the graphs for the supplement. A Meijer carried out the overall analysis of the data and prepared the body of the manuscript. TJ Meerhoff, LE Meeuwen and WJ Paget assisted in the analysis of the epidemiological data. CS Brown assisted in the analysis of the virological data. J van der Velden, as chair person of EISS, contributed by supporting the daily operation of EISS during the 2004-2005 season.

* Members of EISS (during 2004-2005 season):
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- **Austria** - AGES - Institut für Medische Mikrobiologie, Wien; BMGF, Generaldirektion Öffentliche Gesundheit, Wien; Klinikum flor institution für Virologie der Medizinischen Universität Wien, Wien
- **Belgium** - Scientific Institute of Public Health, Brussels
- **The Czech Republic** - National Institute of Public Health, Prague
- **Denmark** - Statens Serum Institut, Copenhagen
- **Finland** - National Public Health Institute, Helsinkı
- **France** - GROG/Open Rome, Paris; Institut Pasteur; Paris; Hospices Civils de Lyon, Lyon
- **Germany** - ArbeitsGemeinschaft Influenza, Marburg; Robert Koch Institute, Berlin
- **Ireland** - National Disease Surveillance Centre, Dublin; Irish College of General Practitioners, Dublin; University College Dublin, Dublin
- **Italy** - Università degli Studi di Milano, Milan; Università di Genova, Genoa; Istituto Superiore di Sanità, Rome
- **Latvia** - State Public Health Agency, Riga
- **Lithuania** - Centre for Communicable Diseases Prevention and Control, Vilnius; Lithuanian AIDS Centre Laboratory, Vilnius
- **Luxembourg** - Laboratoire National de Sante, Luxembourg
- **Malta** - Disease Surveillance Unit, Msida; St. Luke’s Hospital, G’Mangla
- **The Netherlands** - Erasmus University, Rotterdam; Netherlands Institute for Health Services Research, Utrecht; National Institute for Public Health and the Environment, Bithoven
- **Norway** - Norwegian Institute of Public Health, Oslo
- **Poland** - National Institute of Hygiene, Warsaw
- **Portugal** - Instituto Nacional de Saúde, Lisboa
- **Romania** - Cantacuzino Institute, Bucharest
- **Slovakia** - Public Health Authority of the Slovak Republic, Bratislava
- **Slovenia** - National Institute of Public Health, Ljubljana
- **Spain** - Instituto de Salud Carlos III, Madrid; Dirección General de Salud Publica y Consumo, Madrid; Hospital Clinic, Barcelona; Facultad de Medicina, Valladolid
- **Sweden** - Swedish Institute for Infectious Disease Control, Solna
- **Switzerland** - Swiss Federal Office of Public Health, Bern; Laboratoire Central de Virologie, Geneva
- **United Kingdom** - Royal College of General Practitioners, Birmingham, England; Health Protection Agency, London, England; Health Protection Scotland, Glasgow, Scotland; Gartnavel General Hospital, Glasgow, Scotland; NPHS Communicable Disease Surveillance Centre, Cardiff, Wales; University Hospital of Wales, Cardiff, Wales; Communicable Disease Surveillance Centre (NI), Belfast, Northern Ireland; Royal Victoria Hospital, Belfast, Northern Ireland

**Article Supplement available at:**

http://www.eiss.org/documents/eurosurveillance_supplement_2004-2005.pdf for i) movies showing the spread of influenza across Europe, ii) graphs of the weekly consultation rates and virus detections by country, and iii) tables with a detailed breakdown by country of the virological data from sentinel and non-sentinel sources.
Currently, the surveillance of infectious disease in the European Union (EU) is supported by the Basic Surveillance Network (BSN) and other disease-specific surveillance networks (DSNs). Each network has its own website. The objective of the current study was to describe the information presented with public access on each website from the perspective of its usefulness for the surveillance of an EU member state. The BSN and the DSNs cited in Decision 2003/542/CE were included. Each website was reviewed and assessed on the inclusion of characteristics from three broad categories: 1) general information, 2) procedures for data collection and 3) data presentation. Ten surveillance network websites were reviewed during the week of 5 December 2005. At least 80% of the 10 networks included a list of participating countries, the contact addresses for the coordinator of the network and the participating country gatekeepers and the network’s objectives. Only one network specified the source and coverage of the data of each country on its website, and seven presented the disease case definition. Raw data were shown on eight websites and only two networks included presentation of elaborated data for the whole of the EU. Four networks included no reports on their websites. The periodicity of presentation for both raw data and elaborated data varied greatly between networks. The publicly available information on the 10 network websites studied was not homogeneous. We recommend that all networks present a basic set of characteristics on their websites, including case definitions, procedures used for data collection and periodic reports covering elaborated data for the entire EU.

Results

A total of ten networks (BSN and 9 DSNs) and their websites were included in the study [TABLE 1]. Twenty three characteristics were identified: seven characteristics for the category of general information, eight characteristics for the category of procedures of data collection and four for the category of data presentation. The category for data presentation was divided into sections for raw data and elaborated data and four characteristics were disaggregated for each of these sections. The characteristics of the networks’ websites are shown in Table 2.

Introduction

In 1998 the European Union (EU) created an epidemiological surveillance network for the control of infectious diseases covering all EU member states [1]. The following year the list of diseases included by this network was published [2]. The EU-wide network is currently supported by the Basic Surveillance Network (BSN), and other disease-specific networks (DSNs) for the control of infectious diseases [3].

The recent dramatic increase in the use of the internet has facilitated communication within the EU, and epidemiological surveillance networks are therefore increasingly developing the use of the internet to share information, address issues rapidly and communicate to a larger audience. The BSN and each of the DSNs have developed their own websites which allow member states to access disease specific information easily as well as surveillance data from both inside and outside the EU.

The evaluation of websites for the quality of information they present is a growing field and various guidelines exist for this purpose [4-7]. The Health Summit Working Groups have identified criteria for the assessment of the quality of internet health information, these include credibility, content, disclosure, links, design, interactivity and caveats [4]. However, these criteria apply more specifically to websites which share information on health problems, treatment and their prevention. The evaluations of websites relating to surveillance networks are less common and criteria for this purpose are currently not standardised.

The objective of the current study was to describe and compare the information presented with public access on the websites of the BSN and DSNs, from the perspective of usefulness for the surveillance activities of an EU member state.

Methods

The BSN and the DSNs specified in the EU decision 2003/542/EU were included in the study. The European Antimicrobial Resistance Surveillance System (EARSS) was excluded, as it does not address a specific disease but rather a health problem.

It was necessary to identify characteristics of the websites that are considered useful from a member state’s perspective. The identified characteristics were grouped into three broad categories, including: 1) general information, 2) procedures for data collection and 3) data presentation. Within the category of data presentation, raw data were defined as data that had not yet been subjected to analysis. Elaborated data were defined as data presented as reports with some text for their interpretation (not raw data or figures). The websites for each of the networks were then located and examined for these characteristics. Websites were reviewed during the week of 5 December 2005.

General information

Seven networks indicated that they had the participation of all 25 EU countries. All ten networks also included non-EU countries among their members. On all 10 websites reviewed, the participating countries were listed. The contact addresses for the network coordination were presented on nine websites and the contact address for the gatekeepers of participating country on eight websites. Five websites had restricted access links for network members/participating countries only. The principles of collaboration on which the networks are founded were only accessible on four of the websites. All networks presented their objectives on their websites.

Procedures for data collection

The availability of the procedures used by networks for data collection varied across the websites. All networks indicate the diseases under surveillance and, except for the BSN with 49 and ENIVD with 17, the range was between one and four. One network (EISS) specified the source and coverage of the surveillance data for...
each of the participating countries. This network was also the only one to obtain aggregated data by week rather than individual case counts. Over 50% of the networks included sections on the official case definition used and the list of variables collected by the network (5 networks). EuroTB is the only DSN to specify the format used and type of data collected, by making the questionnaires for data collection available to the public. The periodicity with which participating countries sent their surveillance data was specified on five websites and the handbook for procedures followed by the participating countries and the networks on six websites.

**Data presentation: raw data versus elaborated data**

Two networks showed raw data for the entire EU (EuroHIV and EuroTB) and six networks showed raw data for all participating countries combined. Eight networks showed raw surveillance data for each of the participating countries. Two networks for surveillance in Europe (Enter-net and EUVAC.NET) presented only elaborated data. The raw data that were presented by networks was considered to be provisional data in all cases, as they were not indicated as being final data. The periodicity with which raw data are presented on the websites varies by network. EISS, for example, presented raw data for each epidemiological week, and EUROCJD and EuroTB posted raw annual data series. On four network websites, users could request raw data by categories such as country and period.

Four networks (BSN, ENIVD, EWGLINET and EUROCJD) did not show reports with elaborated data on their websites. EuroHIV and EuroTB included specific sections on the EU and surveillance data from the member states of the EU in their reports. The other four networks had elaborated aggregated data for all participating countries and separated by participating country (with the exception

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**Table 1**

Surveillance networks included in the assessment, with their respective abbreviations, diseases surveyed and website addresses (December 2005)

<table>
<thead>
<tr>
<th>Number</th>
<th>Surveillance Network</th>
<th>Abbreviation</th>
<th>Disease</th>
<th>Internet site address</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Basic Surveillance Network</td>
<td>BSN</td>
<td>49 diseases under surveillance for EU</td>
<td><a href="http://www.eubsn.org">www.eubsn.org</a></td>
</tr>
<tr>
<td>2</td>
<td>European Influenza Surveillance Scheme</td>
<td>EISS</td>
<td>Influenza</td>
<td><a href="http://www.eiss.org">www.eiss.org</a></td>
</tr>
<tr>
<td>3</td>
<td>European Network for Diagnostics of “Imported” Viral Diseases</td>
<td>ENIVD</td>
<td>Imported viral haemorrhagic diseases</td>
<td><a href="http://www.enivd.de">www.enivd.de</a></td>
</tr>
<tr>
<td>4</td>
<td>A Surveillance Community Network for Vaccine-preventable Infectious Diseases</td>
<td>EUVAC.NET</td>
<td>Measles, pertussis, rubella, mumps</td>
<td><a href="http://www.sst.dk/euvac/">www.sst.dk/euvac/</a></td>
</tr>
<tr>
<td>5</td>
<td>European Surveillance Scheme for Travel Associated Legionnaire’s Disease</td>
<td>EWGLINET</td>
<td>Travel associated Legionnaire’s Disease</td>
<td><a href="http://www.evgit.org/ewglinet.htm">http://www.evgit.org/ewglinet.htm</a></td>
</tr>
<tr>
<td>6</td>
<td>HIV/AIDS Surveillance In Europe</td>
<td>EuroHIV</td>
<td>HIV and AIDS</td>
<td><a href="http://www.eurohiv.org">www.eurohiv.org</a></td>
</tr>
<tr>
<td>7</td>
<td>International surveillance network for the enteric Infections Salmonella and VTEC O157</td>
<td>Enter-net</td>
<td>Enterohaemorrhagic E. Coli and Salmonella</td>
<td><a href="http://www.hpa.org.uk/hpa/Inter-enter-net_menu.htm">www.hpa.org.uk/hpa/Inter-enter-net_menu.htm</a></td>
</tr>
<tr>
<td>8</td>
<td>European Union Invasive Bacterial Infections Surveillance Network</td>
<td>EU-IBIS</td>
<td>Haemophilus Influenza Group B and Neisseria meningitidis</td>
<td><a href="http://www.euibis.org">www.euibis.org</a></td>
</tr>
<tr>
<td>9</td>
<td>The European and Allied Countries Collaborative Study Group of CJD plus the Extended European Collaborative Study Group of CJD</td>
<td>EUROCJD NEUROCJD</td>
<td>Infectious spongiform encephalopathy, Creutzfeldt-Jakob variant</td>
<td><a href="http://www.eurocjd.ed.ac.uk">www.eurocjd.ed.ac.uk</a></td>
</tr>
<tr>
<td>10</td>
<td>Surveillance of Tuberculosis In Europe</td>
<td>EuroTB</td>
<td>Tuberculosis</td>
<td><a href="http://www.eurotb.org">www.eurotb.org</a></td>
</tr>
</tbody>
</table>

**Table 2 (1)**

Characteristics of the websites of European epidemiological surveillance networks (December 2005)

<table>
<thead>
<tr>
<th>Characteristics studied</th>
<th>Epidemiological surveillance networks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total</td>
</tr>
<tr>
<td>Number of EU participating countries</td>
<td>10</td>
</tr>
<tr>
<td>Number of non-EU participating countries</td>
<td>10</td>
</tr>
<tr>
<td>Contact address coordination</td>
<td>9</td>
</tr>
<tr>
<td>Contact address participating countries</td>
<td>8</td>
</tr>
<tr>
<td>Restricted access link</td>
<td>5</td>
</tr>
<tr>
<td>Principles of collaboration</td>
<td>4</td>
</tr>
<tr>
<td>Objectives</td>
<td>10</td>
</tr>
<tr>
<td>Number of diseases under surveillance</td>
<td>10</td>
</tr>
<tr>
<td>Data source by country</td>
<td>1</td>
</tr>
<tr>
<td>Coverage of data by country</td>
<td>1</td>
</tr>
<tr>
<td>Case definition</td>
<td>7</td>
</tr>
<tr>
<td>List of variables collected</td>
<td>5</td>
</tr>
<tr>
<td>Structure and coding for collected variables</td>
<td>1</td>
</tr>
<tr>
<td>Periodicity with which data is sent to network</td>
<td>5</td>
</tr>
<tr>
<td>Handbook for procedures</td>
<td>6</td>
</tr>
</tbody>
</table>

* Indicates when characteristic was present on network website
of Enter-net). As with the raw data, it was not specified whether data used for reports are final data; however, when annual reports were presented, the elaborated data were considered to be final data. The periodicity with which reports are published varied greatly for all DSNs. Instant reports, such as alert messages, were posted by EISS, Enter-net and EWGLINET. The date of the last report published was also assessed and only EISS and Enter-net had reports relating to surveillance data from 2005.

**Discussion**

Information sharing by EU DSNs through posting on their websites is extremely valuable given that it is a quick and easy way to distribute and access relevant data and information. For member states to fully understand and make use of this information presented on the internet it is important that the websites clearly state the objectives of the network, which diseases are under surveillance and their case definitions, how to contact the network coordinators and members, how data are collected in each country, and that data are presented in a comprehensive manner.

This study tried to assess whether these criteria were addressed by the information presented on the websites of 10 EU surveillance networks [FIGURE]. Of the seven characteristics pertaining to general information, five were fulfilled by more than 80% of the studied networks, which is highly acceptable. Unfortunately, in terms of the data collection procedures, there were several aspects which are insufficiently explored and not homogeneous between the websites. These include: a) how data are obtained by each network and b) the information available for identifying the sources and coverage of both the raw and elaborated data shown. This makes the data presented difficult to use, compare and interpret.

The networks were created to support communicable disease surveillance in the EU, and it is therefore essential that in addition to raw data, all networks include reports on the disease situation in the entire EU and, if possible, for the groups of countries with similar procedures for collecting surveillance data. As shown in the results, only two networks included reports with this information in their websites at the time of the study. The inclusion of such reports would facilitate the comparison of the situation of each disease between

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**Table 2 (II)**

Characteristics of the websites of European epidemiological surveillance networks. (December 2005)

<table>
<thead>
<tr>
<th>Characteristics studied</th>
<th>Epidemiological surveillance networks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total</td>
</tr>
<tr>
<td><strong>Raw Data</strong></td>
<td></td>
</tr>
<tr>
<td>Systematic presentation of raw data:</td>
<td></td>
</tr>
<tr>
<td>A) Data format:</td>
<td></td>
</tr>
<tr>
<td>- For entire EU</td>
<td>2</td>
</tr>
<tr>
<td>- For all participating countries</td>
<td>6</td>
</tr>
<tr>
<td>- By participating country</td>
<td>8</td>
</tr>
<tr>
<td>B) Data consolidation:</td>
<td></td>
</tr>
<tr>
<td>- Provisional</td>
<td>7</td>
</tr>
<tr>
<td>- Final</td>
<td>4</td>
</tr>
<tr>
<td>C) Periodicity:</td>
<td></td>
</tr>
<tr>
<td>- Weekly</td>
<td>1</td>
</tr>
<tr>
<td>- Monthly</td>
<td>0</td>
</tr>
<tr>
<td>- 3-monthly</td>
<td>1</td>
</tr>
<tr>
<td>- 6-monthly</td>
<td>0</td>
</tr>
<tr>
<td>- Annual</td>
<td>4</td>
</tr>
<tr>
<td>- Series of years</td>
<td>5</td>
</tr>
<tr>
<td><strong>Non-systematic presentation of raw data</strong></td>
<td>4</td>
</tr>
<tr>
<td><strong>Elaborated Data</strong></td>
<td></td>
</tr>
<tr>
<td>Systematic reports on the disease:</td>
<td></td>
</tr>
<tr>
<td>A) Data format:</td>
<td></td>
</tr>
<tr>
<td>- For entire EU</td>
<td>2</td>
</tr>
<tr>
<td>- For all participating countries</td>
<td>6</td>
</tr>
<tr>
<td>- By participating country</td>
<td>5</td>
</tr>
<tr>
<td>B) Data consolidation:</td>
<td></td>
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<tr>
<td>- Provisional</td>
<td>2</td>
</tr>
<tr>
<td>- Final</td>
<td>4</td>
</tr>
<tr>
<td>C) Periodicity:</td>
<td></td>
</tr>
<tr>
<td>- Weekly</td>
<td>1</td>
</tr>
<tr>
<td>- Monthly</td>
<td>0</td>
</tr>
<tr>
<td>- 3-monthly</td>
<td>1</td>
</tr>
<tr>
<td>- 6-monthly</td>
<td>1</td>
</tr>
<tr>
<td>- Annual</td>
<td>4</td>
</tr>
<tr>
<td>- Series of years</td>
<td>4</td>
</tr>
<tr>
<td><strong>Non-systematic reports on the disease</strong></td>
<td>3</td>
</tr>
</tbody>
</table>
| **Indicates when characteristic was present on network website**
| **Elaborated data = data which are presented as reports with some text for their interpretation**
Recommendations

Contents of the EU networks’ websites should be reviewed to include a basic set of characteristics that are common to each of these sites. These basic characteristics could include: 1) case definitions, 2) procedures used for data collection and 3) periodic reports which include elaborated data for the entire EU and, if it is possible, also raw data. As the European Centre for Disease Prevention and Control (ECDC) will have a role in harmonising the functioning of the European surveillance networks, it should also take a leading role in establishing guidelines for the inclusion of these basic characteristics on the networks’ websites.

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References

2. European Commission Decision Nº 2000/96/EU, 22 December 1999, lists the diseases that should progressively become covered by the Community network, in application of the European Council and Parliament Decision Nº 2119/98/ EU.

Outbreak report

Two clusters of human infection with influenza A/H5N1 virus in the Republic of Azerbaijan, February–March 2006

Following the appearance of influenza A/H5 virus infection in several wild and domestic bird species in the Republic of Azerbaijan in February 2006, two clusters of potential human avian influenza due to A/H5N1 (HAI) cases were detected and reported by the Ministry of Health (MoH) to the World Health Organization (WHO) Regional Office for Europe during the first two weeks of March 2006. On 15 March 2006, WHO led an international team, including infection control, clinical management, epidemiology, laboratory, and communications experts, to support the MoH in investigation and response activities. As a result of active surveillance, 22 individuals, including six deaths,
were evaluated for HAI and associated risk infections in six districts. The investigations revealed eight cases with influenza A/H5N1 virus infection confirmed by a WHO Collaborating Centre for Influenza and one probable case for which samples were not available. The cases were in two unrelated clusters in Salyan (seven laboratory confirmed cases, including four deaths) and Tarter districts (one confirmed case and one probable case, both fatal). Close contact with and de-feathering of infected wild swans was considered to be the most plausible source of exposure to influenza A/H5N1 virus in the Salyan cluster, although difficulties in eliciting information were encountered during the investigation, because of the illegality of some of the activities that might have led to the exposures (hunting and trading in wild birds and their products). These cases constitute the first outbreak worldwide where wild birds were the most likely source of influenza A/H5N1 virus infection in humans. The rapid mobilisation of resources to contain the spread of influenza A/H5 in the two districts was achieved through collaboration between the MoH, WHO and its international partners. Control activities were supported by the establishment of a field laboratory with real-time polymerase chain reaction (RT-PCR) capacity to detect influenza A/H5 virus. Daily door-to-door surveillance undertaken in the two affected districts made it unlikely that human cases of influenza A/H5N1 virus infection remained undetected.

Introduction

Following anecdotal reports of die-offs of birds in January 2006, influenza A/H5N1 virus infection was confirmed in February 2006 by the State Veterinary Laboratory in Baku in samples obtained from wild birds, commercial poultry (chickens), and backyard poultry (ducks) in central and south Azerbaijan [1]. However, there was reportedly no extensive spread through backyard poultry in the villages. The Republic of Azerbaijan, with approximately 8.4 million inhabitants [2], lies on the shore of the Caspian Sea in the Caucasus, bordering the Russian Federation, Georgia, Armenia, Turkey, and Iran. As common in the whole subregion, migratory birds fly through Azerbaijan twice each year, from Siberia to Africa in the autumn (August-December) and back in the spring (February-May) [3].

On 6 March 2006, the Ministry of Health (MoH) of the Republic of Azerbaijan reported to the World Health Organization (WHO) Regional Office for Europe a cluster of nine cases, including two deaths, of potential human influenza with influenza A/H5N1 HAI [4]. The patients had become ill over a two week period, with dates of illness onset from 15 February to 4 March 2006, and lived in Daikyand settlement in Salyan district, 130 km southeast of the capital, Baku [FIGURE]. Their symptoms included fever, headache, cough and meningeal signs. The clinical presentation was varied, which may have obscured and delayed the suspicion of influenza A/H5N1 virus infection.

On 9 March 2006 another pair of cases where influenza A/H5N1 virus infection was suspected was reported to the MoH from Bayim-Sarov, Tarter district in central eastern Azerbaijan. The date of illness onset of the first case was 28 February 2006 and was initially diagnosed with reactivation of tuberculosis (TB). Because of this diagnosis, influenza A/H5N1 virus infection was only considered when the second patient became ill on 4 March 2006.

On 15 March 2006, further to a request for assistance by the MoH, a WHO-led international team that had been in Azerbaijan since 5 March 2006 to support the implementation of the national surveillance system for HAI was strengthened by experts in infection control, clinical management, epidemiology, laboratory work and communication. The team, which eventually comprised 11 individuals representing five institutions and organisations (Robert Koch-Institut, Germany; Státní Zdravotní Ústav – Centrum Epidemiologie a Mikrobiologie, Czech Republic; US NAMRU-3, Egypt; WHO Headquarters, Switzerland; WHO Regional Office for Europe, Denmark), was deployed in order to assist in

- describing the outbreak;
- public health surveillance, including active case-finding;
- timely and accurate laboratory diagnosis of influenza A/H5 virus infection;
- safe and effective case management and transport of patients for whom influenza A/H5N1 virus infection was considered.

**Figure**

Cluster of human infection with influenza A/H5N1 virus, Daikyand settlement, Salyan District, Azerbaijan, February-March 2006

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Key words: Azerbaijan, Influenza a virus, H5N1 subtype, communicable diseases, emerging, disease outbreaks
Methods

Surveillance system, case finding and case investigation

In accordance with national ministerial decrees issued early in 2006, district chief doctors implemented reporting of cases where influenza A/H5N1 virus infection was suspected from the local doctors to the MoH and informed and trained healthcare workers on how to detect and report such cases. All reported cases were investigated at district level and, after reporting to the central level, also by the MoH-WHO response team.

Since early February 2006, the general public was also informed, through social mobilisation campaigns (e.g. distribution of posters, school lessons) at district and national level, about the risk of exposure to and mode of transmission of influenza A/H5N1 virus, symptoms of AI, and was invited to seek medical care if suggestive symptoms developed.

Daily active surveillance for human cases of influenza A/H5N1 virus infection began on 1 March 2006 in Daikyand settlement. A total of four brigades, each comprising three local healthcare workers, made daily visits to all households (200 households per brigade) to screen residents for fever or respiratory symptoms, through interviews and direct observation. Surveillance data were reported daily by the chief district doctors to the MoH. A similar system became operational in Tarter district around mid-March 2006.

The surveillance team, which included members from the MoH, the Anti-Plague Station (APS), the Republican Centre of Sanitary Hygiene (both technical institutions reporting to the MoH), and WHO, developed a case definition [see Box] and a standardised case investigation form for potential HAI cases, including the following sections: reporting and demographic details, clinical presentation and evolution, history of admission to healthcare facilities, assumption of antiviral drugs as prophylactic or/and treatment measures, history of exposure to animal and human cases, laboratory test results for influenza A/H5N1 virus, final disposition. Both the case definition and the case investigation form were translated into the languages used locally (Azeri and Russian) and used across the country. Data on cases were gathered from multiple sources, including medical records, district medical officers and epidemiologists and directly by interview from family members. When necessary, the interviewing was repeated to collect all the relevant information as further intelligence came to light.

Forms for the monitoring of healthcare workers and workers in the veterinary sector, as well as for contact persons, were developed and an Epi Info 2000 database was created in English and Russian for data entry and analysis.

Laboratory methods

National laboratory capacity was established using a portable field laboratory was established at the APS premises in Baku by the United States Naval Medical Research Unit 3 (NAMRU-3), Cairo (Egypt). The field laboratory included real-time polymerase chain reaction (RT-PCR) with capacity to detect influenza A/H5 virus. All clinical specimens were tested for the presence of influenza A/H5 virus infection using a two-step procedure, involving testing for ‘flu A (matrix gene)’ followed by a second round for H5. No serological tests were performed at the field laboratory.

Regardless of the results obtained in the field, all specimens were transferred to the WHO Collaborating Centre for Influenza at the National Institute for Medical Research (NIMR), Mill Hill (United Kingdom) for confirmation by RT-PCR for influenza A/H5 virus (Asian lineage), haemagglutination inhibition test, virus isolation in embryonated eggs and MDCK cells, and genomic sequencing.

Results

Epidemiology

Cluster 1

Daikyand is a rural, relatively poor village in Salyan district, with around 4800 inhabitants in 800 households. The village is divided in three settlements: Seydler, Daikyand and Salvan [FIGURE].

Influenza A/H5 infection was laboratory confirmed in samples from seven residents of Daikyand settlement. Six were from the same family and one from a neighbouring family, and became ill over a two week period, with dates of onset from 15 February to 4 March 2006. Four of the seven cases died, and this figure is compatible with the case fatality rate observed elsewhere [5]. The median interval between onset of symptoms and death was 9 days (mean: 11.2 days; range: 8–19 days). Patients’ ages ranged from 10 to 20 years (mean: 16 years; median: 17 years); five of the seven cases were females aged 15–20 years.

During the initial interviews, family members denied any contact with sick or dead wild birds or domestic poultry. However, other community members indicated that in February 2006 a massive die-off of swans had occurred in the area and that the family might have had contact with the swans. Following further repeated interviews, relatives of the cases finally revealed that, in February 2006, the family had been involved in de-feathering dead wild swans.

Among the seven cases, the signs and symptoms reported included fever (six), pneumonia (six) cough (five), sore throat (four), shortness of breath (one), stomach pain (one), body aches (one) and meningeal signs (one). All seven cases were admitted to healthcare facilities in Baku during the course of their illness; four were isolated in designated facilities.

Cluster 2

On 28 February 2006, a 24 year old male resident in a camp for internally displaced persons in Bayim-Sarow, Tarter district, in central eastern Azerbaijan, developed shortness of breath, weakness, headache, and had a low grade fever (37.5 °C). As his clinical condition deteriorated, he was admitted to hospital. The patient died on 3 March 2006 with diagnosis of reactivated TB. No samples were conserved for examination and the patient was retrospectively classified as a probable case.

On 4 March 2006, his 18 year old sister developed similar symptoms. On 9 March 2006, three days after referral to Baku, she died, with a diagnosis of TB. However, because of the rapid course of her illness, HAI was suspected. Blood obtained post-mortem tested positive for influenza A/H5 virus infection by the NAMRU-3 field laboratory. These findings were later confirmed at NIMR, Mill Hill (United Kingdom).

In February 2006, a die-off of wild birds had been observed in Tarter district, with no reports of sick poultry in the area. Family members denied that the two siblings had been exposed to sick or dead domestic or wild birds. Information provided by community
members, however, suggested that the siblings had purchased a dead turkey that was thought to have been ill, and then de-feathered it, prepared it and ate it.

**Other districts**
The MoH-WHO team visited other districts identified as being at risk for HAI because of reports of die-offs of birds or laboratory confirmation of influenza A/H5 virus infection in wild birds or poultry. A total of 22 individuals, including six deaths, were investigated for HAI in six districts (Khachmaz, Neftchala, Tarter, Sabail, Salyan and Surakhana) and admitted to healthcare facilities. The final case classification includes eight confirmed cases and one probable case. Of the remaining 13 patients for whom HAI was considered, 12 tested negative for influenza A/H5N1 virus infection and one, from whom no samples were obtained, was diagnosed with another condition following thorough clinical assessment.

**Laboratory**
One hundred and eight clinical specimens (throat and nasal swabs, sera, and rectal swabs) obtained from 20 individuals, in whom a diagnosis of influenza A/H5N1 virus infection was considered and from 32 of their contacts were tested by RT-PCR.

The field laboratory detected seven cases of influenza A/H5 virus infection and NIMR confirmed eight cases. Of the three specimens (throat swabs) that tested negative at the field laboratory and positive at NIMR, two were from patients from whom additional specimens were obtained and subsequently tested positive at the field laboratory. The throat swabs were taken very early in the course of their illness and the viral load was likely to be low. These results are compatible with the lower sensitivity of tests performed by the field laboratory compared to that of tests performed at the WHO Collaborating Centre for Influenza. All positive results obtained by the field laboratory were confirmed by NIMR. No specimens from contacts of patients tested positive for influenza A/H5 virus infection.

Virus strains were isolated from three cases from the cluster in Salyan district. Phylogenetic comparison of H5 haemagglutinins at the WHO Collaborating Centre for Influenza shows that all genes were of avian virus origin and closely related to the sequences of the corresponding genes of other ‘Qinghai Lake’ H5N1 viruses isolated from avian species (including viruses isolated from a swan in Azerbaijan in February 2006, A/swan/Italy/179/06, and from a swan in the Islamic Republic of Iran, A/swan/Iran/754/2006, and from humans (in Turkey, Iraq and Egypt)) [6,7]. These viruses were thus distinguishable from the H5 haemagglutinin of viruses isolated in East Asian countries, including China, Indonesia and Vietnam.

**Case Management**
Clinical care at the regional level is limited in Azerbaijan, and mechanical ventilation is generally not available at district hospitals. Therefore, 16 individuals for whom diagnosis of influenza A/H5N1 virus infection was considered were transferred to three designated AI referral hospitals in Baku. Patients fulfilling the definition of a probable case following the clinical assessment were admitted to an isolation unit at one of these hospitals. Probable and confirmed cases received oseltamivir (150 mg/day for 5 days), antibiotic and critical care support as needed. Severe cases were given oseltamivir up to 10 days, in accordance with WHO advice [8]. Contacts of confirmed and probable cases, including healthcare workers, were subject to health monitoring by the surveillance teams or in the referral hospitals for seven days after the date of their last known contact. None of the contact persons monitored developed symptoms compatible with HAI.

**Discussion**
Between February and March 2006, two clusters of HAI with nine cases (eight confirmed and one probable) were identified in Azerbaijan.

The majority of patients developed respiratory symptoms, with the exception of one patient where meningeval signs were predominant, as already observed in Vietnam [9]. Severe hypoxia, caused by the prolonged course of viral pneumonia, appeared to be under-recognised and treated late in children. The early establishment of oxygen saturation monitoring and provision of continuous oxygen therapy is therefore crucial to prevent decompensation and multi-organ failure already observed in cases of influenza A/H5N1 infection elsewhere [10].

Close contact with and de-feathering of infected wild swans were the most plausible exposures to influenza A/H5N1 virus in the Daikyand cluster, although the investigation of the possible source of infection was made difficult because hunting and trading wild birds and their products is illegal, and therefore there was some reluctance in the community affected to disclose information on possible exposures.Repeated interviews of relatives of the cases finally revealed that, in February 2006, all cases had been involved in de-feathering dead wild swans, after a massive die-off of swans had occurred in the area. Swan feathers are used for pillows and can be sold at a good price in the locality. De-feathering birds is often undertaken by women, which explains the predominance of female cases [10,11].

The HAI cluster in Daikyand settlement is the first event where wild birds were the most likely source of influenza A/H5N1 virus infection in humans. However, the difficulties in gathering accurate information, confusion over reported dates of illness onset, and similar experiences with past influenza A/H5N1 outbreaks where multiple plausible exposures were reported, mean that the possibility that limited human-to-human transmission cannot be ruled out.

The economic implications associated with the ban of hunting and trading in wild birds introduced in October 2006, and the fact that the issue of financial compensation related to potential culling of backyard poultry was not addressed in messages to the population may have hindered effective collaboration with the community. Unfortunately, this might have influenced the implementation of control measures as well as the investigation of the source of infection. However, because of door-to-door surveillance undertaken in Salyan and Tarter districts, it is unlikely that additional HAI cases remained undetected.

The rapid establishment of the RT-PCR laboratory in Azerbaijan provided timely and reliable diagnosis of influenza A/H5 virus infection close to the outbreak site overcoming the difficulties of shipping procedures to NIMR for confirmation which were not well established and subject to delay. The specificity of the field laboratory RT-PCR was supported by the absence of false-positive results. The rapid mobilisation of resources to contain the spread of influenza A/H5 in the two districts was possible because of the close and transparent collaboration between the MoH, WHO and its international partners.

The risk of spread of HAI to western European countries by wild birds is considered to be limited due to widespread awareness that sick and dead wild birds are a potential source of influenza A/H5 virus infection [12].

**Note:** this manuscript has been adapted from the following WER publication: Human avian influenza in Azerbaijan, February-March 2006. Wkly Epidemiol Rec. 2005;81(18):183-8. http://www.who.int/wer/2006/wer8118.pdf

**References**
A need for national guidelines relating to interactive water features was highlighted following three outbreaks of cryptosporidiosis in the United Kingdom, all of which were related to public water features. In August 2003 the Health Protection Agency South West of England was notified of an outbreak of cryptosporidiosis associated with an interactive water feature designed for water play within an adventure park. The water feature was implicated following samples with a high coliform count and the presence of faecal coliforms. A case was defined as any child (younger than 16 years of age) who had visited the park during August and who subsequently had gastrointestinal symptoms and a faecal sample positive for cryptosporidium. Seventy one children were identified in the cohort. This outbreak of cryptosporidiosis was characterised by a very high attack rate (89%), relatively severe in duration (median 8 days) and had a relatively high hospital admission (16% of cases). The epidemic curve was consistent with a point source of infection, and had a relatively high hospital admission (16% of cases). The outbreak report

Method

The cohort population included all children (aged less than 16 years) among household members or friends of a probable or confirmed case who had visited the park with a case during August 2003. A probable case was defined as any child who had visited the park during August 2003 and who subsequently had gastrointestinal symptoms including diarrhoea, blood in stools, vomiting, nausea, or abdominal pain. A confirmed case was defined as a probable case who had visited the park with a case during August 2003 and who subsequently had gastrointestinal symptoms including diarrhoea, blood in stools, vomiting, nausea, or abdominal pain. A confirmed case who had visited the park during August 2003 and who subsequently had gastrointestinal symptoms included diarrhoea, blood in stools, vomiting, nausea, or abdominal pain.

Methods

The cohort population included all children (aged less than 16 years) among household members or friends of a probable or confirmed case who had visited the park with a case during August 2003. A probable case was defined as any child who had visited the park during August 2003 and who subsequently had gastrointestinal symptoms including diarrhoea, blood in stools, vomiting, nausea, or abdominal pain. A confirmed case was defined as a probable case who had visited the park during August 2003 and who subsequently had gastrointestinal symptoms including diarrhoea, blood in stools, vomiting, nausea, or abdominal pain. A confirmed case who had visited the park during August 2003 and who subsequently had gastrointestinal symptoms included diarrhoea, blood in stools, vomiting, nausea, or abdominal pain.

Ten-litre grab samples were taken from the various water features within the park for cryptosporidium oocyst detection by South West Water Ltd. Faecal samples from the farmyard animals were also submitted. Oocysts were detected by light microscopy. Positive specimens were sent to the HPA Cryptosporidium Reference Laboratory for genotyping.
Results

Ninety one children were identified in the cohort, of whom 71 were contacted, giving a 78% response rate. Sixty three children (89%) met the case definition (27 confirmed and 36 probable cases). The sex distribution was even. Median age was 6 years (range 1-15). The most common symptom was diarrhoea (94%), followed by vomiting (64%), abdominal pain (62%), and nausea (51%). None of the children reported blood in stools. The median duration of illness was 8 days (range 1-18) and more than 30% of the children were still ill at the time of interview. Ten children (16%) required hospital admission.

Forty-six of the children who were cases (73%) had visited the park on 8 August, the date of symptom onset for the first case. Of the 51 children whose date of illness onset was known, 45 (88%) had a date of onset within 8 August, the date of symptom onset for the first case. Dates of onset were between 8 and 29 August, and the outbreak peaked on 13 and 14 August. For two of the four cases with date of onset more than 10 days after visiting the park, other household members had had gastrointestinal symptoms in the 10 days before onset. The two probable cases with onset date on date of visit became ill during the evening after leaving the adventure park.

The exposure yielding the strongest association with illness was contact with the interactive water feature [TABLE] (RR= 1.8, CI 95% 0.45 to 7.31, p=0.06). No specific type of contact with this source of illness was significantly associated with illness. This feature involved being sprayed with recirculated water. Children often entered the feature fully clothed and with their shoes on. Nineteen children drank the recycled water and one parent reported that the water ‘smelt like drains’. The filtration and disinfection systems were not adequate to cope with high levels of contamination, and the water feature was closed on 21 August, soon after the start of this investigation.

Samples from 23 of the 27 confirmed cases were sent for genotyping. Sixteen yielded a result and 14 of these were Cryptosporidium parvum genotype 2. The initial sample from the interactive water feature contained a single oocyst that could not be genotyped. Although a subsequent sample from this feature when not in operation was positive and identified as Cryptosporidium parvum genotype 2, there was insufficient DNA for subtyping. Due to a failure of communication, faecal samples taken from animals resident in the park were not tested for cryptosporidium.

Discussion

This outbreak of cryptosporidiosis was characterised by a high attack rate (89% in the cohort studied), long duration of illness (median 8 days) and high proportion admitted to hospital (16%). The dates of onset were consistent with a common source of infection from an exposure in the adventure park. The analytical study showed an association between exposure to water in the interactive water feature and illness. Although the strength of the evidence was reduced due to the small numbers in the unexposed group, the finding was supported by the microbiological results and environmental observations. No association with other water sources or animal contact was detected. It seems likely that water in the interactive water feature became contaminated with faeces containing cryptosporidium oocysts, either from the footwear of users or from an unidentified primary case. These oocysts then continued to circulate in a viable condition as a result of ineffective filtration and disinfection.

In response to the outbreak, the park reviewed and revised health and safety risk assessments to manage and control the risk from protozoan parasites. The design of the water treatment and disinfection system was improved. The park also provided additional drinking fountains around the park and asked children to remove footwear before entering the interactive water feature. They improved signage, instructing visitors at all water-related attractions not to drink the water.

This outbreak has similarities to two others reported in England in 2003 involving public water features. The first, which also occurred in southwest England, involved four cases of cryptosporidiosis in children who had played in a fountain.

Table

<table>
<thead>
<tr>
<th>Exposure</th>
<th>Risk among exposed</th>
<th>Risk among not exposed</th>
<th>Risk Ratio (CI 95%)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Animal contact</td>
<td>36/40</td>
<td>27/31</td>
<td>1.03 (0.87 to 1.23)</td>
<td>0.7</td>
</tr>
<tr>
<td>Small animal petting area</td>
<td>20/22</td>
<td>10/11</td>
<td>1.00 (0.80 to 1.23)</td>
<td>0.3</td>
</tr>
<tr>
<td>Small animal and reptile handling</td>
<td>8/8</td>
<td>18/20</td>
<td>1.11 (0.96 to 1.27)</td>
<td>1.0</td>
</tr>
<tr>
<td>Farm animal petting area</td>
<td>24/27</td>
<td>5/5</td>
<td>0.89 (0.78 to 1.02)</td>
<td>1.0</td>
</tr>
<tr>
<td>Young farm animal petting area</td>
<td>6/7</td>
<td>20/21</td>
<td>0.30 (0.65 to 1.23)</td>
<td>0.0</td>
</tr>
<tr>
<td>Water contact in park</td>
<td>63/71</td>
<td>0 Not exposed</td>
<td>Not calculated</td>
<td>Not calculated</td>
</tr>
<tr>
<td>Log flume</td>
<td>34/42</td>
<td>27/29</td>
<td>0.92 (0.79 to 1.08)</td>
<td>0.3</td>
</tr>
<tr>
<td>Boats</td>
<td>14/16</td>
<td>48/54</td>
<td>0.98 (0.80 to 1.21)</td>
<td>0.9</td>
</tr>
<tr>
<td>Interactive feature</td>
<td>62/68</td>
<td>1/2</td>
<td>1.82 (0.45 to 7.31)</td>
<td>0.06</td>
</tr>
<tr>
<td>River walk</td>
<td>12/13</td>
<td>48/54</td>
<td>1.04 (0.86 to 1.25)</td>
<td>0.7</td>
</tr>
<tr>
<td>Contacts in Log flume</td>
<td>14/16</td>
<td>17/21</td>
<td>1.08 (0.82 to 1.43)</td>
<td>0.7</td>
</tr>
<tr>
<td>Hand/Face</td>
<td>20/25</td>
<td>7/7</td>
<td>0.8 (0.66 to 0.97)</td>
<td>0.6</td>
</tr>
<tr>
<td>Body Only</td>
<td>4/4</td>
<td>24/29</td>
<td>1.21 (1.02 to 1.43)</td>
<td>1.0</td>
</tr>
<tr>
<td>Contacts in Interactive feature</td>
<td>54/59</td>
<td>1/1</td>
<td>0.92 (0.45 to 0.99)</td>
<td>0.8</td>
</tr>
<tr>
<td>Hand/Face</td>
<td>15/16</td>
<td>11/11</td>
<td>0.94 (0.83 to 1.06)</td>
<td>0.4</td>
</tr>
<tr>
<td>Body only</td>
<td>18/19</td>
<td>30/34</td>
<td>1.07 (0.91 to 1.26)</td>
<td>0.4</td>
</tr>
</tbody>
</table>
The water feature comprised two separate water bodies with separate holding tanks and water treatment systems using bromide and sand filtration. A large pool with water to a depth of 20cm was used as a paddling pool, although it was not intended for this purpose. Cryptosporidium oocysts were isolated from all four cases and detected in water samples taken from the fountain.

The second outbreak, which occurred in central England, was linked to a newly opened purpose-built interactive water feature, and involved 122 cases. More than 80% (102) of those infected were under 15 years old. Thirty five (85%) of 41 cases tested for cryptosporidium were positive. Indicator organisms of faecal contamination were identified from the water but no cryptosporidium oocysts were recovered.

These outbreaks raised issues about the lack of national guidance on operation and maintenance of water-based recreational attractions, which have now been addressed by the United Kingdom Pool Water Treatment Advisory Group [8]. The principal public health measure for preventing infections and outbreaks associated with these devices is risk assessment and management. The principal microbiological risks are cryptosporidiosis from inadequate filtration, and bacterial and viral infections, including legionella, from inadequate disinfection. This guidance proposes design and operational standards for filtration, chlorination and reducing contamination hazards.

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References


Outbreak report

Outbreak report

OUTBREAK OF E. coli O157 INFECTION IN THE SOUTH WEST OF THE UK: RISKS FROM STREAMS CROSSING SEASIDE BEACHES

C Ihekweazu1, M Barlow1, S Roberts1, H Christensen1, B Outridge1, D Lewis1, S Paynter1,2

In August 2004 seven cases of Escherichia coli O157 infection were identified in children on holiday in Cornwall, southwest England, all of whom had stayed at different sites in the area. Isolates from all seven cases were confirmed as E. coli serogroup O157 phage type 21/28. We carried out a case-control study among holidaymakers who visited the beach. A standardised questionnaire was administered by telephone to parents. They were asked where on the beach the children had played, whether they had had contact with the stream that flowed across the beach, and about their use of food-outlets and sources of food eaten. Cases were more likely to have played in the stream than controls (OR [1.72- undefined]). The time spent in the stream by cases was twice spent there by controls. Cases and controls were equally exposed to other suspected risk factors. PFGE profiles for all the cases were indistinguishable. Increased numbers of coliforms were found in the stream prior to the outbreak. Cattle were found grazing upstream. We suggest that the vehicle of infection for an outbreak of acute gastrointestinal illness caused by E. coli O157 was a contaminated freshwater stream flowing across a seaside beach. The onset dates were consistent with a point source. Heavy rainfall in the days preceding the outbreak might have lead to faeces from the cattle potentially contaminated by E. coli O157 contaminating the stream, thereby leading to the outbreak. Control measures included fencing off the part of the stream in which children played, and putting up warning signs around the beach.

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Key words: beaches, case-control, E. coli, outbreak, O157, UK

Introduction

Human infection with verocytotoxic Escherichia coli O157:H7 (E. coli O157) is associated with clinical illness ranging from non-bloody diarrhoea to haemolytic uraemic syndrome (HUS) and death. It is the most common cause of renal failure in children [1,2]. It is transmitted to humans through contaminated food, water, and direct contact with infected people or animals [2-4]. The infectious dose is very low, under 100 organisms [2,5]. E. coli O157 is one of the most commonly identified causes (25% in 2002) of recreational fresh water-associated outbreaks involving gastroenteritis in the United States [6].

In August 2004 seven cases of E. coli O157 infection were identified in children who had been on holiday in Cornwall (resident population 500 000), a popular holiday destination in southwest England. Initial investigations found that the patients had been camping at different sites but had all played in a stream flowing across the same beach within a period of a few days. Isolates from all seven cases were

References

confirmed as *E. coli* serogroup O157 phage type 21/28. The pulsed field gel electrophoresis (PFGE) profiles for all the isolates were indistinguishable. Prior to this there had been no clustering of this phage type in the area. The other six cases of *E. coli* O157 reported via the surveillance system with links to Cornwall during August were phage types 2 or 8. None of the patients in these six cases had visited this beach. We carried out a case-control study to search for supportive evidence that the stream or other exposure was the vehicle of infection in this outbreak of *E. coli* serogroup O157 phage type 21/28.

**Methods**

Cases were defined as children aged between one and ten years, with laboratory confirmed *E. coli* O157 phage type 21/28 infection, present at the beach at any time between 11 and 18 August 2004, and with onset of illness between two and eight days after a visit to the beach. This definition included all cases of confirmed *E. coli* O157 phage type 21/28 infection. Two of the seven cases were siblings, and so to avoid introducing potential bias arising from common behaviour patterns found in sibling groups, random numbers were used to choose one of these two children. This resulted in six cases for the study.

Controls were defined as children aged between one and ten years who visited the same beach between 11 and 18 of August (the range of likely exposure dates of the cases). We considered that the source population of cases consisted of all tourists who had stayed in various campsites in the area and we set out to recruit four controls per case from residents of holiday campsites in this area.

The primary hypothesis was that cases were more likely than controls to have been exposed to water from the stream. Also, the time spent playing in the stream by cases and controls was compared. The use of local food outlets and restaurants, and types of food consumed, were also investigated.

A detailed standardised questionnaire was administered by telephone to parents of cases and controls. Cases and controls were asked where on the beach they sat or played (each was sent a map and so to avoid introducing potential bias arising from common behaviour patterns found in sibling groups, random numbers were used to choose one of these two children. This resulted in six cases for the study.

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The primary hypothesis was that cases were more likely than controls to have been exposed to water from the stream. Also, the time spent playing in the stream by cases and controls was compared. The use of local food outlets and restaurants, and types of food consumed, were also investigated.

A detailed standardised questionnaire was administered by telephone to parents of cases and controls. Cases and controls were asked where on the beach they sat or played (each was sent a map of the local area to mark areas where the children had played and eaten), whether they went in the stream, used food outlets, used toilet facilities and washed hands before eating. The questionnaires were entered into an EpiData (v 3.02) database. STATA (v 8.2) was used to analyse the data.

Environmental investigation included sampling water from the stream and cattle grazing on the surrounding fields above the stream. Stream sediment samples were tested using immunomagnetic enrichment.

**Results**

**Epidemiological Investigation**

All seven cases had laboratory confirmed *E. coli* O157 phage type 21/28. They were all very ill, with clinical symptoms including diarrhoea, abdominal pain, vomiting and blood in stool, and four required admission to hospital. All played in the same stream for some time between 7 and 23 August 2004 (six on 15 August 2004). Six of the seven cases definitely had contact with the stream on the three days between 15 and 17 August 2004 [TABLE 1].

**Six of the seven cases were included in the case-control study.**

Four hundred and twenty families were contacted by phone from lists of residents from four local campsites. We identified 27 children who were eligible as controls. The ages of the six cases ranged from 3 – 7 years with a median of 5.5 years. The ages of the controls ranged from 1 – 10 years with a median of 7. The mean age was 5.2 years for cases and 6.7 for controls. (p = 0.191). Males and females were equally distributed between cases and controls (p=0.665).

Cases were more likely to have played in the stream than controls (OR [1.72- undefined]).

p = 0.02) [TABLE 2]. Of the children who played in the stream, cases were more likely to have had water splashed onto their faces than controls (OR [1.22- undefined], p = 0.05) [TABLE 2]. The time spent in the stream by cases was twice that spent by controls. This difference was not statistically significant, but there was a dose response using the mid-point of time played in stream as the exposure score (p= 0.002) [TABLE 3]. Cases and controls were equally exposed to each of the other suspected risk factors.

**Table 2**

<table>
<thead>
<tr>
<th>Variable</th>
<th>Cases (n=6) of <em>E. coli</em> O157 phage type 21/28 and controls (n=27) according to possible exposure factors, Cornwall, United Kingdom, August 2004</th>
</tr>
</thead>
<tbody>
<tr>
<td>Played in stream</td>
<td>6 (100.0)</td>
</tr>
<tr>
<td>Played in the sea</td>
<td>6 (100.0)</td>
</tr>
<tr>
<td>Played around pipes</td>
<td>1 (16.7)</td>
</tr>
<tr>
<td>Consumed food/drink</td>
<td>4 (66.7)</td>
</tr>
<tr>
<td>Bought food/ drink for child</td>
<td>4 (66.7)</td>
</tr>
<tr>
<td>Brought own food / picnic</td>
<td>4 (66.7)</td>
</tr>
</tbody>
</table>

Further analysis of those who did play in the stream

| Variable | Cases Controls OR (95% CI) P-value |
|----------|----------------------------------|-------------------------------|
| Average time in stream (hours) | 3.17 1.58 0.10 |
| Splashed water onto face | 6 (100.0) | 6 (50.0) | 1.22 (0.47, 3.53) | 0.05 |
| Lay flat in stream water | 2 (33.3) | 1 (8.3) | 5.5 (0.20, 353.18) | 0.25 |
| Sat in stream water | 6 (100.0) | 7 (58.3) | 0.88 (0.02, 4.17) | 0.11 |
| Washed hands in stream | 3 (50.0) | 11 (91.7) | 0.09 (0.002, 1.87) | 0.08 |
| Drank water from stream | 2 (33.3) | 0 (0.0) | 1.20 (0.17, 9.00) | 0.10 |

**Table 3**

<table>
<thead>
<tr>
<th>Cases of <em>E. coli</em> O157 phage type 21/28 and controls according to time spent in the stream, Cornwall, United Kingdom, August 2004</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time spent playing</td>
</tr>
<tr>
<td>----------------------</td>
</tr>
<tr>
<td>No time</td>
</tr>
<tr>
<td>Less than 1 hour</td>
</tr>
<tr>
<td>1 - 2 hours</td>
</tr>
<tr>
<td>3 - 4 hours</td>
</tr>
<tr>
<td>&gt; 4 hours</td>
</tr>
<tr>
<td>Total</td>
</tr>
</tbody>
</table>

* Chi square for linear trend = 9.70 p = 0.00184

**Environmental Investigation**

After the incident, environmental samples positive for *E. coli* O157 were found at five sites, both in the stream and in cattle faeces in the catchment area of the stream. None of the environmental isolates were phage type 21/28.
Routine sampling in the stream had recorded an increase in contamination from total and faecal coliforms in the lower reaches of the stream in the two months before the outbreak, but there were no tests carried out specifically for E. coli serogroup O157. Three potential sources of sewage contamination from overflow drains around the stream were discovered upstream.

Discussion

This investigation supports the hypothesis that the vehicle of infection for an outbreak of acute gastrointestinal illness caused by E. coli O157 was a contaminated freshwater stream flowing across a seaside beach in Cornwall. The illness onset dates and the dates of contact with the stream are consistent with a point source. In 1999, a similar outbreak involving E. coli O157 phage type 21/28, associated with a bathing beach, occurred in a neighbouring county [7], but this is the first reported outbreak of E. coli O157 in the UK associated with recreational exposure to a stream.

The exact source contamination of the stream was not discovered. E. coli phage type 21/28 was not detected, despite extensive sampling of cattle faeces. However, this remains the most likely source of contamination of the stream. Cattle are a major reservoir for human infection with E. coli O157. Previous studies have suggested that shedding by animals is seasonal and that people with greater exposure to livestock are at a greater risk of infection [8, 9]. In the two days preceding the outbreak, heavy rainfall was recorded locally: 200.4 mm of rain fell in 24 hours on 16 August [10], leading to severe flooding [11]. This may have increased the likelihood of cattle faeces contaminated by E. coli O157 being washed into the stream. Another potential source for contamination of the stream was sewage overflow from overflow drains around the stream.

Initial control measures included fencing off the lower part of the stream in which children played, and putting up signs to warn people of the potential dangers of contact with the stream. The media also helped to inform residents and visitors of the area of the potential danger of playing in the stream. In the long term, a series of multi-agency meetings were initiated to assess the potential risks from such streams. This led to the initiation of environmental studies of the effects of summer storms on the bacteriological quality of local streams in 2005. Evidence from this study will inform future public health policy.

There is substantial potential for contamination of streams flowing across beaches and leading to outbreaks such as the one described above, especially in areas with a high cattle population. Rainfall and run-off have been implicated in outbreaks of E. coli O157 in the past [12, 13] and the use of weather monitoring and forecast information has been used in the United States to predict day-to-day water quality for beach advisories [14]. This outbreak highlights the importance of E. coli O157 as a waterborne pathogen with a low infective dose, which allows water to act as an efficient vector [15]. We recommend that efforts are made to increase public awareness of this potential hazard and to explore with the agricultural industry other methods of reducing faecal contamination of streams and rivers, especially those used for recreation.

Acknowledgements

We would like to acknowledge the contributions of Geraldine Smith and Tom Cheasty from the HPA’s Laboratory of Enteric Pathogens, Richard Bendall from Royal Cornwall Hospital, the Environment Agency, Restormel Borough Council, SouthWest Water, and the Veterinary Laboratories Agency.

References

Two outbreaks of measles in Germany 2005

A Siedler1, A Tischer2, A Mankertz2, S Santibanez2

Measles re-emerged in some counties in Germany in 2005, despite increasing vaccination coverage rates in children at school entry in recent years, which had led to decreasing incidences (with the lowest incidence ever recorded, 0.2 cases per 100 000 inhabitants in 2004). Regional outbreaks have been detected by the mandatory reporting system in the states of Hesse and Bavaria. Although both outbreaks led to similar incidences in the affected areas (14 and 12 cases respectively per 100 000 inhabitants) they differed in age distribution, transmission patterns and measles virus genotype.

In Hesse, 223 cases were submitted, from which 160 belonged to 41 clusters mainly defined by family or household contacts. Attack rate was highest in children aged between 1-4 years (102 cases per 100 000). Results of measles virus diagnosis showed genotype D4 and identical nucleotide sequences for all analysed cases from Hesse. In Bavaria, 279 cases were submitted, most of which had occurred in schools and preschool facilities. Age-specific attack rate was highest in children aged between 5-9 years (129 per 100 000). Results of measles virus diagnosis showed genotype D6 and identical nucleotide level. In both outbreaks the vast majority of cases (95% in Hesse and 98% in Bavaria) were in unvaccinated children, but vaccination coverage differed in the affected areas and was slightly lower in Bavaria than in Hesse. Local accumulation of unvaccinated children and their concentration in schools and kindergarten preceded the outbreak in Bavaria. Despite high average vaccination coverage levels, local variations may lead to regionally limited outbreaks.

Methods
Both outbreaks were detected by the mandatory reporting system which is based on the Protection Against Infection law (“Infektionsschutzgesetz”) [4]. According to this law, physicians must report every suspected measles case, and laboratories must report every confirmed measles case, to the local health department. At the local level, which consists of 431 county health departments nationwide, reports are checked to see whether they fit the case definition, whether clinical and laboratory reports may be linked, and whether further cases have occurred which have not been reported yet. Case data are electronically submitted to the health departments of Germany’s 16 federal states and from there to the RKI. Cases are listed according to the reporting week, which is given by data entry at local level.

Each measles case submitted must meet one of the three following diagnostic categories:
- Clinically diagnosed case: fever and rash and at least one of the symptoms cough, coryza, conjunctivitis, Koplik spots
- Clinically and laboratory confirmed case: clinically diagnosed case with laboratory confirmation
- Clinically and epidemiologically confirmed case: clinically diagnosed case without laboratory confirmation but with an epidemiological link to a laboratory confirmed case

In the following report a case is defined as any submitted case, regardless of diagnostic category, unless another explanation is given.

Local health authorities carried out outbreak investigations by interviewing physicians and family members in order to detect further cases and contacts. In order to stop transmission they began campaigns in schools and kindergartens, aimed at informing parents and getting susceptible children vaccinated by their family physicians.

After detection of the first contact cases, the federal health authorities, together with the NRC MMR, encouraged public health officials and physicians in the affected areas to carry out laboratory investigations. Tests were carried out in local private laboratories and in the NRC MMR. Local laboratories generally test sera for measles specific IgM and IgG antibodies by commercially available enzyme immunoassays. Information on the total number of tested but not confirmed suspected measles cases is available only from the NRC MMR.

In the NRC MMR antibody tests were carried out as well as detection of MV RNA in clinical samples (throat swabs, urine and oral fluid) by RT-PCR, as described previously [5]. In order to trace the transmission pathways of the virus, samples from 38 cases were observed by sentinel and mandatory surveillance in the western part of the country [1,2]. Only sporadic cases occurred in the eastern part (territory of the former German Democratic Republic) due to higher vaccination coverage [1,2]. Since 2003, the incidence of reported cases nationwide has dropped below 1 per 100 000 inhabitants [3]. Vaccination coverage registered at school entry has steadily increased from 89% and 15% (for the first and second dose, respectively) in 1998 to 94% and 66% in 2004. However, there are differences in vaccination coverage at regional and local levels. At the beginning of 2005 two measles outbreaks were detected by the surveillance system in counties of the federal states of Hesse and Bavaria. In this report both outbreaks are described including genetic analysis of the detected MVs in order to illustrate how and why regionally limited outbreaks may still occur.

Introduction
In Germany, two doses of MMR vaccine have been recommended since 1991. The current schedule has been in place since 2001, and recommends that the first dose is given at age 11 to 14 months and the second dose at age 15 to 23 months. Vaccination is mainly done by private physicians. Vaccination coverage and measles control remain regionally different in the federal states. Nationwide measles surveillance started in 1999 with a sentinel group of paediatricians and general practitioners (GPs), which was kept in place when statutory reporting was introduced by law in 2001. Case reports in both systems are made according to the clinical case definition. Laboratory testing of suspected measles is mostly offered and carried out in a decentralised fashion by private laboratories. However, the National Reference Centre for Measles, Mumps and Rubella (NRC MMR) at the Robert Koch-Institut (RKI) plays a major role particularly in genotyping of measles viruses (MVs).

The epidemiological situation has changed in recent years. Until 2002 endemic circulation and regional outbreaks of measles were
Measles outbreaks 2005 in Hesse and Bavaria (Germany):

**Results**

**Outbreaks in Hesse**

From January to May 2005, a total of 223 cases were reported from four neighbouring counties (the cities of Offenbach, Frankfurt, Wetterau and Giessen) and the nearby city of Wiesbaden accounting for an incidence of 14 cases per 100 000 inhabitants in this area. During the same period, a further 29 sporadic cases were reported from 11 counties of Hesse, but 10 counties of this federal state had no measles cases.

Age-specific attack rates were highest in children aged between 1-4 years (102 per 100 000), followed by those aged 5-9 years (83 per 100 000) [FIGURE 1]. Although the incidence in adults was only about two per 100 000, the rate of admission to hospital was 34% in patients aged 20 years and older. A fourteen year old girl died.

The vast majority (n= 209; 95%) of cases were in unvaccinated people.

The first clusters of measles cases were reported in the cities of Offenbach and Frankfurt, mainly in families considered to be hard to reach by the health services. A case report of a hospitalised patient in January led the public health authorities to identify further patients with cases which fit the clinical case definition but who had not seen a physician. Nineteen of the cases reported in January 2005 had experienced onset of disease in 2004.

Measles cases were next reported from the adjacent county of Wetterau, where several families were affected, followed by reports from the county of Giessen and finally from the city of Wiesbaden [FIGURE 2].

One hundred and sixty six cases from the five counties were scattered in 41 clusters with clinically and epidemiologically confirmed cases, mainly defined by family or household contacts. Despite interviews with patients, parents and other carers and guardians, and physicians, no connections between the clusters themselves or between the clusters and the remaining single cases were detected.

A diagnosis of measles was laboratory confirmed in 67 cases. The NRC MMR obtained samples from 29 suspected measles cases in the state of Hesse and confirmed measles diagnosis in 18 cases, all of which were distributed in the five counties affected by the outbreak. Results of MV genotyping available for 12 patients from Hesse showed that these cases were exclusively caused by MVs of the same genotype D4. Moreover, these MVs also showed identical nucleotide sequences and thus belonged to a homogeneous genetic group.

**Outbreak in Bavaria**

From March to July, 279 cases were submitted from eight counties in the south of Bavaria, in and around the city of Munich, leading to an incidence of 12 cases per 100 000 inhabitants in the region [FIGURE 3]. During the same period, 25 sporadic cases were submitted from 13 further Bavarian counties. No cases of measles were reported in the remaining 75 counties.

The outbreak mainly affected school aged children (5-14 years old) (n=208; 74%) but about 12% of cases were in adolescents and adults (n=16), and 7 out of 11 hospitalised cases were in patients aged 20 years or older.

Age-specific incidence was highest in children aged between 5 and 9 years (129 cases per 100 000 children), followed by those aged between 10 and 14 years (58 per 100 000) [FIGURE 1].

As the attack rates indicate, most of the cases were related to outbreaks in schools or preschool facilities: 45 cases occurred in a primary school in Munich, 52 cases in children from several counties who attended the same Montessori school, 42 cases in children in four kindergartens, and 38 cases in four further schools in different communities. Investigations of the local health authorities showed...
possible transmission between these outbreak settings. This was also confirmed by laboratory results.

Seventy of the reported outbreak cases were laboratory confirmed, 26 of these were tested in the NRC MMR, and MVs from 17 cases representing all local clusters were genotyped. All of these viruses were identified as genotype D6 and were identical at the nucleotide level. This indicates the presence of the same chain of transmission of a D6 virus within the Bavarian outbreak.

Most of the cases (n=273; 98%) were in unvaccinated people, including eight children who were initially reported as vaccinated, but vaccine had been given during the incubation period, which was too late to prevent the disease. The genetic identification of four of these cases revealed measles wild-type virus (D6). In six cases, vaccination status remained unknown.

One measles case in Austria could be traced to the Bavarian outbreak, but no information on the genotype was available.

Discussion

Although vaccination coverage seemed to be high on average, regional outbreaks still occurred. In the affected region in Hesse, vaccination coverage at school entry is on the same level as the nationwide average proportion: 95% and 65% for the first and second dose, respectively. This might explain why most of the cases observed where either single cases or part of small clusters. Virus circulation was ultimately limited because vaccinated people were well protected and this led to the interruption of the transmission chain. The age distribution of the cases in Hesse and the peak at age 1-4 years suggest that vaccination is not given at the recommended age which was below two years of age for two doses. Some of the affected families were part of a particular community where most families had several children, avoided seeking medical care, are difficult for healthcare services to reach, and do not bring their babies to healthcare services for routine checkups. Missing vaccinations for the children of such families are usually detected and given later in childhood (for instance at medical examination before school entry), leaving the very young unprotected, and therefore susceptible children may accumulate. Additionally, coverage of the second dose of vaccine is generally still too low to make up for primary vaccine failures and to use the early second chance to be effectively immunised. Unfortunately, vaccine coverage data by age are not available. The registration of coverage at school entry is too late to assess whether children were immunised appropriate to age and to identify target groups for catch up vaccination.

Vaccination coverage in Hesse is slightly higher than in Bavaria (91% and 59% for the first and second doses, respectively) and, moreover, there are great regional and local differences in vaccination coverage in Bavaria. In the affected Bavarian counties, coverage is below the Bavarian average (personal communication, Dr. Hautmann, Bayerisch Landesamt für Gesundheit und Lebensmittelsicherheit). This may explain why it took a longer time for a similar number of people to be infected in a smaller area in Hesse in comparison to Bavaria.

However, the older age of the Bavarian measles patients demonstrated that clusters of unvaccinated people may benefit from herd immunity until the virus arrives. Public health authorities had observed a concentration of unvaccinated children in single communities and certain schools and childcare facilities (most of which had connections with the anthroposophic teachings of Rudolf Steiner) in the outbreak areas in advance but their vaccination recommendations, although publicised in local newspapers and handouts to parents and carers in schools and kindergartens, were apparently ignored. This might have led to the accumulation of measles-susceptible people and the rapid spread of infection.

The virus of the observed transmission chain in Hesse differs from the previously detected D4 viruses. No identical nucleotide sequence could be found in the published data so far. Interestingly, the NRC MMR as the WHO regional reference laboratory had investigated clinical material from eight cases belonging to a measles outbreak in Romania in the fourth quarter of 2004. The detected D4 MV’s share the nucleotide sequence with the D4 viruses which emerged in Hesse in the 1st quarter of 2005 and in Berlin in the 2nd quarter of the same year [FIGURE 4]. Therefore, it can be assumed that the detected D4 MVs in Germany were possibly imported from Romania. This assumption is supported by the public health authorities in Hesse, who informed about possible contacts of cases in Hesse to Romania.

The genotype D6 MVs in Bavaria share their sequence with those of 4 measles cases from Switzerland also investigated at the NRC MMR, which occurred in the first quarter of 2005. Moreover, the only case confirmed by the NRC MMR in 2004 (second quarter, federal state of North-Rhine-Westphalia) belonged to the same variant of genotype D6. During the 1990s, MVs of genotype D6 were not only endemically circulating in Germany but also widely distributed throughout Europe [6,8-11]. Furthermore, sequence data published in the GenBank indicate that the same genetic variant of D6 was also circulating in several regions of Russia in 2003 and 2004. Therefore, the appearance of a D6 virus in Bavaria might be due to a continued limited circulation of this genotype in central Europe or might likewise be caused by virus importation.

Conclusion

The mandatory reporting system already in place enabled health authorities and epidemiologists at all levels of public health to detect and combat outbreaks of measles.

Laboratory investigation plays an important role in measles surveillance and control, and is particularly indispensable for tracing transmission chains in outbreaks. Genetic characterisation of the detected viruses revealed that the outbreaks in Hesse and Bavaria were associated with distinct MV genotypes. These data demonstrate that both outbreaks were caused by independent transmission chains of the MV. While the outbreak in Hesse was possibly due to imported measles, the origin of the Bavarian outbreak could be either imported or indigenous.

Besides the different MV genotypes, the spread of infection also appeared to be different in both outbreaks. While in Hesse, frequent small clusters and single cases were observed in outbreak settings such as families and households, in Bavaria it was mainly childcare facilities where measles susceptible children were concentrated that were affected. It can be assumed that although vaccine coverage was high at average, regional and local variations in vaccination coverage lead to distinct epidemiological situations.

In the two outbreaks two different groups of ‘hard-to-reach’ populations were involved: people who did not generally seek medical care, and people who are selective about the medical services they use and often refuse vaccination, especially for measles. Special attention
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should therefore be given to identifying target groups and to find appropriate ways to reach them by additional immunisation initiatives. This includes assessment of vaccination coverage at an earlier age.

Generally, coverage of the second dose of measles vaccine still needs to be improved at all local, regional and nationwide levels. The outbreaks provide evidence that, despite the decline in measles incidence in Germany due to increased vaccination coverage and improved measles surveillance in recent years, the potential for local outbreaks is still present, and measles control and vaccination awareness should be continued and improved at all levels.

Acknowledgements

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References


Original Articles

Outbreak report

A Regional Outbreak of S. Typhimurium in Denmark and Identification of the Source Using MLVA Typing

M Torp Dahl1, S. G. Sørensen2, S. Ethelberg3, G. Sandøe4, K. Gammelgård1, L. J. Porsbo1

In Denmark, as part of the national laboratory-based surveillance system of human enteric infections, all S. Typhimurium isolates are currently sub-typed using phage typing, antibiogram typing, and pulsed-field gel electrophoresis (PFGE). However, the discriminatory ability of PFGE is not always high enough to discriminate within certain phage types, and it is not always possible to separate unrelated and related isolates. We have therefore applied multiple locus variable number of tandem repeats analysis (MLVA) for surveillance typing of S. Typhimurium since 2004. In May and June 2005, an outbreak with 26 cases of S. Typhimurium infection was identified by MLVA. The isolates were fully sensitive and had one of the most frequently occurring Danish phage types (DT12) and PFGE types. S. Typhimurium DT12 isolates from routine surveillance of animals and food were typed using MLVA and PFGE for comparison with the human isolates. The typing results revealed that an isolate from a pig herd and its corresponding slaughterhouse located in the same geographic region as the outbreak had the same PFGE and MLVA type as the human isolates. In contrast, all other DT12 isolates investigated, which had the same PFGE profile, had different MLVA types. The conclusion that the pig herd was the source of the human infections was supported by patient information, and pork from the herd stopped entering the market on 29 June. MLVA may contribute significantly to both surveillance and outbreak investigations of S. Typhimurium, as without MLVA typing this outbreak would not have been found nor its origin traced.

Introduction

In Denmark there is a large and coordinated surveillance of salmonella infections in food-production animals. Salmonella enterica subspecies enterica serotype Typhimurium (S. Typhimurium) is the second most frequent serotype causing infections in humans after S. Enteritidis [1].

Typing is an important tool for surveillance as well as for investigating outbreaks of human S. Typhimurium infections, and as part of surveillance in Denmark, all S. Typhimurium isolates are routinely typed for resistance, phage, and pulsed-field gel electrophoresis (PFGE). PFGE has been shown to be useful in investigations of S. Typhimurium outbreaks [2,3] and is widely used in local, national and international surveillance [1,4,5]. Unfortunately the discriminatory ability of both PFGE and phage typing is not always high enough within S. Typhimurium when trying to link outbreak isolates. The discriminatory ability of PFGE is particularly low within DT12 and DT104 (two of the most frequent phage types in Denmark) where 80%-90% of all human infections are caused by the same PFGE type. Multiple locus variable number of tandem repeats analysis (MLVA) is a new and promising typing method [6] that has been shown to have good discriminatory power within

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4. EUROSURVEILLANCE VOL.11 Issues 4-6 Apr-Jun 2006
S. Typhimurium and within the uniform phage type DT104 [7]. We have therefore begun using MLVA for routine surveillance of human S. Typhimurium infections.

An outbreak was discovered based on an increased level of a specific MLV A type between 8 May and 23 June 2005. The outbreak included 26 case-patients, four were children under 5 years, three were adults over 70 years, 15 were females, and one case died. Sixteen of the patients lived in the same county.

**Methods**

**Bacterial isolates and phenotypic characterisation**

Isolates were cultured and serotyped using antisera from Statens Serum Institut in accordance with the Kaufman-White scheme [8]. S. Typhimurium isolates were further phage typed at the Danish Institute for Food and Veterinary Research in accordance with international standards [9].

**PFGE procedure**

Isolates were grown overnight on blood plates and PFGE was performed using the PulseNet USA protocol developed for salmonella [5]. The gels were analysed and interpreted using BioNumerics 4.0 (Applied Maths, Sint-Martens-Latem, Belgium). All bands between 33 and 1135 Kb were included in the interpretation of PFGE patterns and isolates differing at one band were assigned a new PFGE type.

**MLVA**

MLVA was performed using the same primers and a modified version of the method previously described [6]. Isolates were grown overnight on blood plates and a small loophole of cells was taken directly into the PCR mix. PCR was performed using a multiplex kit from Qiagen (Hilden, Germany) in a total of 25 μl and including 2.50 pmol of each of the primers STTR3-F, STTR3-R, STTR6-F and STTR6-R and 1.25 pmol of each of the primers STTR5-F, STTR5-R, STTR9-F, STTR9-R, STTR10pl-F and STTR10pl-R. Amplification was performed using a GeneAmp9700 (Applied Biosystems, Foster City, USA), starting with 15 min at 94ºC, followed by 25 cycles of 30 s at 94ºC, 1 min at 60ºC and 1.5 min at 72 ºC and ending with an extension step for 10 min at 72ºC. Fragment sizes for all loci were imported to BioNumerics 4.0 and allele numbers were assigned for each strain. Unique allelic combinations were assigned a new MLVA type.

**Case definition and case-control study**

Cases were defined as S. Typhimurium positive with a distinct MLVA type with onset of disease prior to the intervention at 29 June 2005. Based on initial hypothesis-generating patient interviews a case-control study was conducted, beginning on 21 June. Controls were selected from the Danish population register, matched by municipality, sex, and week of birth. Participants were interviewed by phone using a questionnaire focusing on consumption of a number of varieties of pork and beef, besides other types of meat, fruit, vegetables, places where food was bought, and other exposures.

**Results**

Since June 2004, MLVA typing has been used for routine surveillance of human S. Typhimurium infections in Denmark. In the beginning of June 2005, a cluster of isolates with the same MLVA type (JPX.0216.DK) was found. The isolates were phage typed to DT12 and all isolates also had identical PFGE types. During the time of the outbreak 26 isolates with this particular MLVA type were found in humans over a period of seven weeks [FIGURE 1B]. Figure 2 shows the distribution of human MLVA types within DT12 isolates with the most frequently seen PFGE type (PFGE22) of all Danish human isolates from June 2004 to June 2005. Most MLVA types contained between one and three isolates and only three major clusters of MLVA types, JPX.0216.DK, JPX.0052.DK and JPX.0056.DK were found within the period. Two of the MLVA types, JPX.0052.DK and JPX.0056.DK resulted in human outbreaks in the summer of 2004 and the new cluster, JPX.0216.DK, therefore also seemed to be caused by a common source [FIGURE 2].

**Figure 1**

S. Typhimurium infections with the epitype, Denmark, 2005

**1A:** S. Typhimurium outbreak cases by the date of onset of symptoms (n=26*)

**1B:** All human cases with the epitype in Denmark in 2005, by week of sample receipt at the laboratory

**Figure 2**

Distribution of MLVA types within S. Typhimurium DT12 isolates with the most common PFGE type, Denmark

Note: Distribution of MLVA types in human isolates that were typed as part of national surveillance from June 2004– June 2005. Distribution of MLVA types in food and animal isolates that were typed as part of the outbreak investigation, MLVA types are shown for three clusters containing an increased number of human isolates

Geographical assessment of the cases showed that the majority lived within the same region of the county of Funen and the investigation focused on a local source. The regional veterinary and food control authorities were notified and a local slaughterhouse, from which a sample positive for DT12 had recently been obtained, was identified. Pigs from a local pig herd with a history of clinical illness were slaughtered on the same day that the isolate was found positive for DT12, and isolates from both the slaughterhouse and the pig herd were typed with MLVA. To get an idea of the diversity of MLVA types from different animal and food sources, 13 other isolates originating mostly from pork sampled at slaughterhouses during the outbreak period were included in the analyses. Furthermore, 21 isolates that
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had previously been typed were also included. The distribution of MLVA types within DT12 isolates with the most frequently seen PFGE type from animal and food sources is shown in figure 2. From the total number of 36 food and animal isolates, only two were found to have an MLVA type identical to the outbreak type, namely the isolate from the abovementioned local pig herd and the isolate from the slaughterhouse where pigs from this herd had been slaughtered. The diversity within the rest of the isolates was high and the isolates were separated into 20 different MLVA types (Figure 2). An isolate from the sow herd that delivered pigs to the local pig herd was also included in the investigation and the isolate differed from the outbreak type by one PFGE band, however the MLVA profile was identical.

Concomitant with the microbiological investigation, a case-control study was conducted. It comprised 21 patients and 82 controls. No specific type of food, nor any shop or supermarket was particularly prevalent among cases or found to be associated with disease in matched or unmatched analyses. However, 19 patients reported possible consumption of pork prior to falling ill and 20 patients reported consumption of beef. Almost all cases appeared to have been infected locally.

After the discovery, on 29 June, that the specific herd was the suspected source of the outbreak, pork from this herd was taken off the market and a press statement was released by the Danish Veterinary and Food Administration. No further patients were identified during a three week period following this intervention, but this was followed by a second cluster of nine patients in a six week period (Figure 1B), the majority of whom also lived in the same geographical region. Patient interviews indicated that these patients were not infected via the pig herd that had been identified, and a continued investigation by typing was undertaken under the hypothesis that these cases were acquired faster and at a lower cost and MLVA data were also easier to analyse and interpret. The standardisation of MLVA makes it possible to exchange data between laboratories and we routinely exchange data between Denmark and Norway either as fragment sizes or allelic combinations. We also found that MLVA is a highly discriminatory method and we were clearly able to discriminate between DT12 isolates with the most common PFGE type [FIGURE 1]. In conclusion, we found that MLVA is a highly useful method for surveillance and outbreak investigations of S. Typhimurium.

Discussion

In 2004, DT12 was the most common phage type within S. Typhimurium accounting for 18% of human S. Typhimurium infections in Denmark [1]. PFGE has been used for surveillance of S. Typhimurium isolates and several clusters of PFGE types as well as tracking of common source outbreaks have successfully been done. Unfortunately, discrimination within DT12 and therefore cluster detection is difficult with PFGE; in Denmark we find that 80% of all DT12 isolates have the same PFGE type. MLVA [6] is currently used for routine surveillance of human S. Typhimurium infections in Norway and has been shown useful in outbreak situations [10]. We therefore started using MLVA for routine surveillance of human S. Typhimurium infections.

An outbreak including 26 patients with S. Typhimurium DT12 was detected by MLVA. The majority of patients lived in a confined geographic region. Isolates from a local pig herd and a local slaughterhouse were also typed and had the same PFGE and MLVA types. PFGE and MLVA typing of other food and animal isolates revealed a high diversity of MLVA types within DT12, whereas all isolates were assigned to the same PFGE type. On this basis, it was concluded that the increase of human infections was caused by pork that originated from a local pig herd processed at the local slaughterhouse. The case-control study was inconclusive, but patient interviews support the conclusion reached by the typing. We suspect that the contaminated pork was used to make a large number of different pork-products, giving the case-control study insufficient power. Eating pork is a very common exposure in Denmark. A second cluster of human isolates with the same PFGE and MLVA type was found three weeks after intervention. It is possible that the continued occurrence of the outbreak type was due to other pig herds receiving pigs from the sow-herd where Salmonella with a different PFGE profile but a identical MLVA profile was isolated. This would allow further spread of the outbreak type, although on a smaller scale.

The increase of human S. Typhimurium isolates might possibly have been discovered using phage typing and PFGE typing, but neither of the two typing methods would have been useful for separating outbreak related and non-related human cases or tracking the source of the outbreak and thus MLVA was the best method for the current outbreak investigation. There were several other advantages of MLVA for routine surveillance when compared with PFGE. Data were acquired faster and at a lower cost and MLVA data were also easier to analyse and interpret. The standardisation of MLVA makes it possible to exchange data between laboratories and we routinely exchange data between Denmark and Norway either as fragment sizes or allelic combinations. We also found that MLVA is a highly discriminatory method and we were clearly able to discriminate between DT12 isolates with the most common PFGE type [FIGURE 1]. In conclusion, we found that MLVA is a highly useful method for surveillance and outbreak investigations of S. Typhimurium.

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References

In May/June 2005 an outbreak of diarrhoeal illness occurred among company employees in Copenhagen. Cases were reported from seven of eight companies that received food from the same catering kitchen. Stool specimens from three patients from two companies were positive for Campylobacter jejuni. We performed a retrospective cohort study among employees exposed to canteen food in the three largest companies to identify the source of the outbreak and to prevent further spread. Using self-administered questionnaires we collected information on disease, days of canteen food eaten and food items consumed. The catering kitchen was inspected and food samples were taken. Questionnaires were returned by 295/348 (85%) employees. Of 247 employees who ate canteen food, 79 were cases, and the attack rate (AR) was 32%. Consuming canteen food on 25 May was associated with illness (AR 75/204, RR=3.2, 95%CI 1.3-8.2). Consumption of chicken salad on this day, but not other types of food, was associated with illness (AR=43/97, RR=2.3, 95%CI 1.3-4.1). Interviews with kitchen staff indicated the likelihood of cross-contamination from raw chicken to the chicken salad during storage. This is the first recognised major Campylobacter outbreak associated with contaminated chicken documented in Denmark. It is plausible that food handling practices contributed to transmission, and awareness of safe food handling and storage has since been raised among kitchen staff. The low number of positive specimens accrued in this outbreak suggests a general underascertainment of adult cases in the laboratory reporting system by a factor of 20.

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## Introduction

Campylobacter species, particularly C. jejuni and C. coli, are important causes of acute bacterial gastroenteritis of varying severity. Symptoms include (occasionally bloody) diarrhoea, abdominal pain, fever, nausea and vomiting. In rare instances the infection is complicated by Guillain-Barré syndrome. Although the infectious dose required for infection is low, most cases are sporadic [1]. In Denmark, Campylobacter is the most frequent cause of bacterial diarrhoea and with an incidence of 69 per 100,000 population in 2004, Campylobacter accounted for more than twice the number of Salmonella episodes [2-3]. Despite this, and in contrast to Salmonella, only one large Danish Campylobacter outbreak has previously been described, a waterborne outbreak that occurred in the mid-1990s [4]. Foodborne Campylobacter outbreaks that have been registered to date in Denmark have been few and included relatively small numbers of people [5-8].

On 6 June 2005, a general practitioner reported a case of Campylobacter gastroenteritis on suspicion of a possible foodborne outbreak. The patient was employed in a company in Copenhagen and had mentioned that other employees had similar illness. On 7 June, the regional public health office in Copenhagen received the notification and alerted the Regional Food Control Authority (RFCA). Initial enquiries revealed that the company canteen received food from a catering kitchen that catered for eight companies, and that diarrhoeal illness had been reported among staff in seven of these eight companies and that many employees had fallen ill around 28-29 May. It was therefore likely that canteen food was implicated in disease transmission, and an outbreak investigation was launched by the RFCA and the Statens Serum Institut (SSI) to identify the vehicle of the outbreak in order to remove the source and to prevent future spread.

## Methods

We did a retrospective cohort study among employees exposed to canteen food in the three largest companies affected (known here as A, B and C). Based on the typical incubation period of campylobacteriosis (2-5 days) [9] and reports of peak incidence on 28 and 29 May, exposure was mostly likely to have occurred between Monday 23 May and Friday 27 May. Self-administered paper questionnaires were distributed to employees on 15 June and information was collected on demographic details, symptoms, time of onset and duration of illness, number of days absent from work, type of healthcare contact, canteen food consumption by day (from 23 May to 3 June) and the individual canteen food items consumed in the canteen on 24 and 25 May. A case was defined as an employee in company A, B or C, who had consumed canteen food between 23 May and 3 June and who developed either diarrhoea (> 3 loose stools/day) or abdominal pain and fever after 23 May.

The RFCA inspected the catering kitchen and interviewed kitchen staff about food handling practices and illness. Processed and unprocessed food specimens were collected on 9 and 13 June and examined by the RFCA. Cases were asked to submit stool samples for standard bacteriological and virological analysis. Positive Campylobacter isolates were speciated by PCR and subtyped by automated ribotyping (Riboprinter; Qualicon) using the restriction enzyme HaeIII.

## Results

Of the 348 employees in companies A, B and C, 295 (85%) returned questionnaires. Of these, 47 people had not been exposed to canteen food during the study period and were therefore excluded. One questionnaire was excluded because outcome information was missing. Therefore, 247 questionnaires were included in the analysis. The median age in this cohort was 39 years (range 20–64 years), and 131 (53%) were male. Seventy nine employees met the case definition. The overall attack rate was 32%. The company-, gender- and age-specific attack rates are shown in Table 1.

Day of illness onset for 77 cases is shown in Figure 1; information on date of onset was missing for two cases. After a slight increase beginning on 26 May, the number of cases rose sharply to a distinct peak on 28 May and decreased then exponentially during the following two weeks. Nine patients provided stool samples [FIGURE 1]. Four samples (three with illness onset on 28 May, one on 29 May) were culture positive for Campylobacter, three of these samples were from employees of company A and one was from company C. One of the four isolates was discarded immediately after culturing in the diagnostic laboratory, leaving three isolates for further typing. These were all found to be C. jejuni and were found to have identical DNA
Illness was of longer duration in early cases (median 4.5 days) than in late cases (median 2 days); and more early cases (42%) than late cases (5%) presented with the three concurrent symptoms of diarrhoea, abdominal pain and fever.

Individuals who had consumed canteen food on 25 May were 9.8 times (95% CI 1.4–68.3) more likely to be an early case than people who were not exposed to canteen food on that day. The relative risk of being an early case after consumption of chicken salad was 3.6 (95% CI 1.6–8.0) [TABLE 3]. For late cases there was no association between consumption of chicken salad and being ill (AR=5/97, RR = 0.7, 95% CI 0.2–2.6). Furthermore, no specific day of canteen food consumption was significantly associated with being a late case.

Telephone interviews with staff in the five other companies that had served food from the catering kitchen revealed that in four companies, at least 6 of a total of 58 employees developed a gastrointestinal illness compatible with the case definition, all of them either on 28 or 29 May. Three cases had eaten chicken salad on 25 May, two could not be interviewed and one did not remember whether or not this item had been eaten. No illness was reported in the three people employed at the fifth company.

Interviews with three out of five kitchen workers revealed that raw chicken had been stored in the refrigerator directly on top of the fried chicken that was later used in the chicken salad, with the result that juices from the raw chicken are likely to have dripped onto the fried chicken. The raw chicken fillets used originated from France. Food specimens from the exposure period were no longer available in the catering kitchen at the time of inspection. However, samples were taken from the chicken fillets available in the kitchen at that time, which was a different batch of chicken from the same wholesaler and the same French producer. These chicken breast fillets tested positive for Campylobacter, but the isolated strain was of a different ribotype than the one isolated from the cases. Because poultry is frequently contaminated with Campylobacter [10], no trace-back was attempted.

Discussion

The results suggest that the vehicle of transmission in this outbreak was chicken salad prepared by the catering kitchen and served to employees of company A, B and C on 25 May. The likely infectious agent was Campylobacter jejuni. This finding is not surprising, given that consumption and handling of poultry is believed to be the primary source of Campylobacter infections in the developed world [11] (a recently published case-control study of sporadic Campylobacter infections in Denmark found fresh chicken to be the main risk factor) [12] and given that outbreaks due to cross contamination of cooked food by raw poultry have been described before [1,13]. Considering the high incidence of Campylobacter infections and the fact that a substantial proportion of retail chickens are known to be contaminated [2], it is surprising, however, that an outbreak like the one described here had not previously been reported in Denmark.
Our study may be limited by recall bias, as data were collected around three weeks after exposure. It is likely that some participants reported food habits rather than food items actually consumed. Therefore the true RR may be higher than the observed. Information on food items was not collected for all potential days of exposure, but there was no indication that exposure took place on days other than 25 May. No food items from the exposure period were available for testing. Exposure to chicken salad was homogeneously distributed among the age groups and cannot explain the lower attack rate in older employees, which does not have a straightforward explanation.

The length of the incubation period, the rarity of secondary Campylobacter infections, the difference in clinical symptoms, and the negative culture results of all cases with late onset of illness that were subsequently found to be positive for Campylobacter jejuni infections in the United States and Other Industrialized Nations. In: Nachamkin I., Blaser M.J., editors. Campylobacter. 2 ed. Washington DC: ASM Press; 2000. p. 139-54.

Data from this outbreak may be used to gain a rough estimate of the relationship between the number of Campylobacter cases registered in the Danish laboratory surveillance system and the true number of cases in the community. Three patients decided to see a physician as a result of their illness and had a faecal sample taken for examination, which were subsequently found to be positive for Campylobacter. The remaining five patients who submitted stool samples did so only when asked by the outbreak investigation team. Therefore, only three positive remaining five patients who submitted stool samples did so only when asked by the outbreak investigation team. Therefore, only three positive

1. 69% (58/85) and 66% (57/86) of all cases with late onset of illness that was positive for Campylobacter jejuni.
2. 26% (21/81) and 30% (24/80) of all cases with late onset of illness that was positive for Campylobacter jejuni.

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MEASLES OUTBREAK IN GERMANY: OVER 1000 CASES NOW REPORTED IN NORDRHEIN WESTFALEN

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The reported number of measles cases linked to the current outbreak in the federal state (Land) of Nordrhein Westfalen, in the west of Germany, has now risen to over 1000 [1]. Between 1 January and 3 May 2006, 1018 cases were notified to the health authorities, and this number is believed to be an underestimate, as some cases are not notified, or are not diagnosed.

In recent weeks, the number of notified measles cases has been stable at around 120-140 cases per week. No significant decrease has yet been observed during the school holidays. Particularly efficient transmission has been noted in the Nordrhein region, where 56 cases of measles per 100 000 inhabitants have been reported in the city of Duisburg, 33 per 100 000 inhabitants in the district of Wesel and 53 per 100 000 inhabitants in the district of Mönchengladbach.

Age distribution
School-age children are still the main group affected, representing over 60% of all reported cases. There have been 252 cases reported in children aged 10-14 years, 198 cases reported in children aged 15-19, and 186 cases reported in children aged 5-9. Sixty four cases in children under one year old have also been reported (Figure).

Vaccination status
The majority of patients have not been vaccinated against measles. According to current data, only 25 patients (2.5%) had received a full course of vaccination against measles (2 doses of measles, mumps and rubella (MMR) vaccine).

Complications
About 15% of the patients required hospital admission. Two cases with serious complications (measles encephalitis) have been reported. Other reported complications include 20 cases of lung infection and 17 middle-ear infections.

Laboratory diagnostics
About one third of cases have been laboratory confirmed by detection of virus-specific antibodies or by PCR. As has already been reported, the outbreak in Nordrhein Westfalen is caused by the D6 measles virus, which is the same strain that is currently causing a large outbreak in the Ukraine [2]. It is not yet known whether there is any link between cases in the two countries. The D4 strain of the virus has been found in samples from two patients, suggesting that there are at least two parallel infection chains in Nordrhein Westfalen.

Current control measures
The Nordrhein Westfalen state public health authority are keeping all local health authorities informed of the situation, and are urging actions to increase vaccination coverage in areas where it is low. All local authorities have been supplied with information for distribution to schools, nurseries, parents and doctors. Questionnaires for use when notifying cases have also been supplied. It has been recommended that patients or their parents/guardians are interview to establish the patients’ likely infection source.

Further recommendations include:
- Implementation of vaccination campaigns: checking of vaccination status of all members of the public, and vaccination offered to those found to be unprotected. Healthcare workers within local communities are being encouraged to offer prophylactic vaccination to all patient contacts.
- Local authorities should contact all schools and nurseries within the affected areas, and distribute information to all teachers, parents, nurseries and pupils.
- 14-day isolation of susceptible members of a household of a measles patient from community settings, with re-introduction after post-exposure vaccination.
- Avoidance of contact with patients with confirmed measles outside the household.
- Informing the local media of the outbreak situation.
- Submitting samples from measles testing to the healthauthorities for testing.

At the invitation of the Nordrhein Westfalen authorities, the Robert Koch-Institut in Berlin has assisted with interviewing 1200 people and determining the vaccination status at a school in Duisburg where there were 37 patients. Current studies aim to determine the contribution of areas of low coverage to the outbreak and vaccination records are being studied. All patients whose records show that they are not protected will receive an information leaflet provided by the Deutsches Grünes Kreuz e.V. (DGK, http://www.dgk.de).

The Nordrhein Westfalen state health authorities are also carrying out a telephone survey of all known patients in Duisburg. This survey will provide data needed to compile comprehensive information on the extent of the outbreak, illness length, possible infection sources and transmission routes.

This article was translated and adapted from reference 1 by the Eurosurveillance editorial team.

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CURRENT LEGIONELLOSIS OUTBREAK WITH 139 CASES IN PAMPLONA, SPAIN

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By 8 June 2006, 139 cases of legionellosis had been reported in an outbreak in Pamplona, north Spain. All cases presented with clinical signs of pneumonia, compatible radiography and positive urinary antigen test. The outbreak was recognised on 1 June, when 4 confirmed cases were reported to the Public Health Institute of Navarra. The number of cases diagnosed up to 8 June are presented in the figure, by date of diagnosis. Seventy six of the patients (55%) were admitted to hospital, and the other sixty three patients have been given treatment to take at home. A total of seven patients have required intensive care, and six patients remained in intensive care on 8 June, two of whom are seriously ill. No deaths have occurred. Men represent 47% of cases. The patients range in age between 21 and 97 years.

Most of the initial cases occurred in a neighbourhood close to the city centre, and the investigations began on 1 June with the inspection of 30 cooling towers in 11 buildings in this part of the city. Rapid tests for Legionella antigen were positive in four of the towers, located in three buildings, on 2 June, and these four towers were shut down immediately. Culture and PCR for Legionella have been positive in two of these cooling towers, but could not be confirmed in the other two. The Public Health Institute in Navarra found Legionella with low bacterial load in two further cooling towers, which were shut down on 6 June. A helicopter inspection of the area was carried out on 2 June and identified eight structures that resembled undeclared cooling towers in the investigated area, but further investigation has found that none of these structures is a cooling tower.

Microbiological culture of respiratory samples from patients are in progress. Legionella isolates from the four positive cooling towers have been sent to the reference laboratory in the National Centre of Microbiology in Majadahonda, Madrid.

The local health authorities have been issuing regular press releases giving the details of the outbreak [1-6].

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FLOODING IN EUROPE: A BRIEF REVIEW OF THE HEALTH RISKS

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In the light of current flooding events in Bulgaria, Serbia and Romania [1], staff at the European Centre for Disease Prevention and Control (ECDC) have undertaken some preliminary review of the adverse health effects of such natural disasters.

Flooding events are the most frequently occurring natural disasters worldwide, and may increase in the future as a result of climate change [2]. Adverse effects on human health include [3,4,5]:
• trauma deaths, mainly by drowning;
• injuries;
• enteric infections due to increased faeco-oral cycling from disruption of sewage disposal and safe drinking water infrastructure;
• mental health such as post-traumatic stress disorder;
• vector borne disease, such as malaria, dengue and dengue hemorrhagic fever, yellow fever, and West Nile fever;
• rodent-borne disease, such as leptospirosis;
• poisoning caused by toxic substances;
• snake bites as snakes tend to seek shelter in households to escape from flooding;
• other negative health outcomes, such as disruption of healthcare services and population displacement.

A limited number of short term epidemiological studies have been undertaken to assess the health impacts of flooding, but there is a deficiency in studies of long term health and economic impacts. Population resilience is likely to vary widely depending upon the economic and organizational resources available.

Limited data on flood events shows that the greatest burden of mortality is from drowning, heart attacks, hypothermia, trauma and vehicle related accidents [4,5]. The speed of onset of floodwaters is a factor determining the number of immediate flood-related deaths.

Flood-related injuries, such as contusions, cuts, sprains have been reported in several studies [5,6], as well as burns, electrocutions, snake bites and wound infections. After the tsunami of December 2004, 106 cases of tetanus and 20 deaths were reported in Indonesia (case-fatality ratio 18.9%) [7]. However, the number of serious injuries observed after violent flooding events generally turns out to be much lower than initial estimates predict.

Figure.
Numbers of legionellosis cases by date of diagnosis, Pamplona, May-June 2006

Microbiology in Majadahonda, Madrid.

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Several studies in developed countries have reported increases in mental health problems such as anxiety, depression, sleeplessness, and post-traumatic stress disorder among flood victims [6]. A recent survey of flooded individuals and a reference group of non-flooded individuals from the same area of residence in the United Kingdom [8] found a fourfold increase in psychological distress among adults whose homes were flooded compared with those whose homes were not (RR=4.1, 95% CI: 2.6-6.4). The risk estimates for physical illness in adults declined after adjustment for psychological distress, while psychological distress remained strongly associated with flooding after adjustment for physical illnesses. Other previous studies reported behaviour change in children as increased bedwetting and aggression [9].

There is some evidence that diarrhoea disease increases after flooding, particularly in developing countries, but also in Europe [6]. A recent UK study reported an increase in self-reported gastroenteritis associated with flooding and with increasing risk the greater the depth of household flooding (RR 1.7 [9.0,3.0] p for trend by flood depth = 0.04) and an increase in earache (RR 2.2 [1.1,4.1]) [7]. The large displacement of population that occurs after flooding, and poor sheltering conditions and crowding may also contribute to increase the risk of diarrhoeal and respiratory infections. Other studies refer to evidence of flood-associated outbreaks of leptospirosis in a wide range of countries, including Portugal (1969), the Russian Federation (1997), and the Czech Republic (2003) [3,6,10]. Transmission is believed to be promoted by skin and mucous membrane contact with water, damp soil, vegetation or mud contaminated with rodent urine. Prompt recognition of the disease and early treatment of cases is essential to minimise the impact of the outbreak.

Floods may lead indirectly to an increase in vectorborne diseases through the expansion in the number and range of vector habitats. Standing water caused by heavy rainfall or overflow of rivers can act as breeding sites for mosquitoes, and therefore enhance the potential for exposure of the disaster affected population and emergency workers to infections such as dengue, malaria and West Nile fever. Flooding may initially flush out mosquito breeding, but this will return when the waters recede. Malaria epidemics in the wake of flooding are a well-known phenomenon in malaria-endemic areas worldwide. West Nile fever has emerged in Europe after heavy rains and flooding, with outbreaks in Romania in 1996-97, in the Czech Republic in 1997 and Italy in 1998 [3]. There is also an increased risk of infection of diseases contracted through direct contact with polluted waters, such as wound infections, dermatitis, conjunctivitis, and ear, nose and throat infections.

The effects in developed regions, such as Europe, may be different to those in developing regions. The World Health Organization Regional Office for Europe has been developing several programmes related to assessing the health effects of climate changes, including flooding, such as the project Climate Change and Adaptation Strategies for Human Health (cCASHh) [11] that covers aspects of impact and adaptation assessment for possible climate-related health outcomes in Europe. The recent Rapid Health Assessment of Flooding in Bulgaria [12], reported in 2005, covers the main public health issues that should be considered during and after a flood and is one of the most consistent documents on assessing the current situation and providing recommendations for local response to flooding.

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HIV transmission in part of the US prison system: implications for Europe

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A study in the United States (US) [1] has shown that HIV transmission has been occurring within the prison system in the state of Georgia. Between July 1988 and February 2005, 88 prisoners tested HIV-antibody negative at mandatory testing on entry to prison, and HIV-antibody positive in a subsequent requested test, indicating seroconversion during incarceration. Risk behaviours in prison, specifically sex between men and tattooing, were associated with HIV seroconversion. The estimated HIV prevalence in the US prison population is 2% [2], and a number of European countries have a considerably higher prison HIV prevalence, in some cases, more than 10% [3]. Considering the high HIV prevalence among prisoners in some European countries, and the limited number of HIV prevention and harm reduction programmes currently in place, the US study highlights the need to address and prevent bloodborne virus transmission among prisoners in Europe [4].

The US study found that those prisoners who had seroconverted to HIV were ten times more likely to report sex between men in prison than matched controls (adjusted odds ratio [AOR] 10.1, p-value<0.01), and fourteen times more likely to have been tattooed while in prison (AOR 13.7, p-value=0.01). To a lesser degree, characteristics also associated with seroconversion in prison were having a body mass index ≤25 kg/m2 on entry to prison (AOR 3.8, p-value=0.02), and being of black race (AOR 3.7, p-value=0.03). Prisoners themselves suggested that HIV prevention in prisons should include condom distribution (38%), HIV education (22%), and safe tattooing practices (13%). The study concluded that this clear evidence of transmission within the prison system indicated that effective HIV prevention is needed in prisons.

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Injecting drug users and prisons in Europe

HIV prevalence in European prisons has been associated with injecting drug use and tattooing [5,6,7,8], but continuing HIV transmission within prisons has never been documented. Similar to other Western countries, injecting drug users are overrepresented among the European prison population [4]. A recent study among drug users in 10 European cities reported that 60% had injected drugs in the past year and 55% had already been imprisoned [9]. Studies indicate that between 8% and 60% of prisoners in Europe have used drugs in prison, including intravenously [10]. In common with the United States, European prisoners are more likely than the general population to be HIV-infected, inject drugs and share injecting equipment if they continue to inject in prison [11]. Imprisonment rates in western Europe are typically 50-100 per 100,000 population [3]. However, in the Russian Federation, the rate is 600 per 100,000 population [3], second only to the US, where the rate is over 700 per 100,000 population [12].

The prevalence of HIV in European prisons varies between less than 1% in England to 11% in Portugal and 12% in Estonia [3]. Together with high rates of imprisonment among injecting drug users, of whom about one half continue to inject in prison, and evidence of other risk behaviours for HIV transmission including sex between men and tattooing, HIV and its prevention in prisons is of considerable importance in Europe. While there is growing evidence that HIV transmission in prisons can be reduced [13], current prison HIV prevention and harm reduction provision within Europe remains scarce and frequently inferior to provision in the community.

Evidence that harm reduction and prevention programmes in prisons are effective

A review of prison-based syringe exchanges in Europe found that, overall, reported drug use decreased or remained stable over time, and that syringe sharing declined dramatically. In addition, no new cases of HIV, hepatitis B or hepatitis C transmission were reported [14]. Despite the evidence supporting the value of prison needle and syringe exchange, Spain is the only European country with a systematic programme [10]. Similarly, other HIV harm reduction measures such as substitution treatment, distribution of disinfectant tablets and condoms and other evidence-based harm reduction programmes are lacking or underdeveloped and uncoordinated in European prisons. The WHO Declaration on Prison Health as Part of Public Health calls for equivalent healthcare provision in prisons and the community [15]. Nonetheless, prison health in many European countries continues to be controversial, with relatively little advocacy for equal health protection among prisoners, many of who represent a number of marginalised populations including injecting drug users and other substance misusers, the homeless, and individuals with complex mental health needs. Controversy over healthcare provision in prisons has proved a challenge to implementing HIV harm reduction strategies, despite increasing recognition that good prison health is good public health. Missing the opportunity to address and prevent HIV transmission in prisons will result in failure to prevent HIV transmission in the community, since most prisoners are eventually released from prison and return to being citizens. The opportunity to prevent infectious disease, including HIV, in both prisons and the community is a significant and frequently unrecognised element of public health protection.

Conclusions

The demonstration of HIV transmission in prisons in part of the US highlights the following implications for European prisons:

- The need for prison-specific advocacy and commitment on the political and public health agendas.
- The need to include prison staff in all stages of prevention and harm reduction.
- The need for joint efforts by all professionals working in prisons, decision makers (such as the relevant government ministries, prison administrations, and nongovernmental organisations) and international bodies (such as WHO and the United Nations Office on Drugs and Crime) to address infectious disease prevention in prisons.
- The need to adapt and introduce into prisons harm reduction approaches proven to be cost-effective and efficient in the community.

More information on drugs and infections in European prisons can be found at http://www.endipp.net, the website of the European Network on Drugs and Infections Prevention in Prison (ENDIPP), ENDIPP is a Europe-wide, multidisciplinary network that is active in all 25 EU member states and accession countries, and co-funded by the European Commission’s Public Health Programme.

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