Recurrent outbreaks of *Salmonella* Enteritidis infections in a Texas restaurant: phage type 4 arrives in the United States

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(Accepted 16 February 1996)

SUMMARY

In recent years infection caused by *Salmonella* serotype Enteritidis (SE) phage type 4 has spread through Europe but has been uncommon in the USA. The first recognized outbreak of this strain in the USA occurred in a Chinese restaurant in El Paso, Texas, in April 1993; no source was identified. In September 1993, a second outbreak caused by SE phage type 4 was associated with the same restaurant. To determine the cause of the second outbreak, we compared food exposures of the 19 patients with that of two control groups. Egg rolls were the only item significantly associated with illness in both analyses (first control group: odds ratio [OR] 8.2, 95% confidence interval [CI] 2.3–31.6; second control group: OR 13.1, 95% CI 2.1–97.0). Retrospective analysis of the April outbreak also implicated egg rolls (OR 32.4, 95% CI 9.1–126.6). Egg roll batter was made from pooled shell eggs and was left at room temperature throughout the day. These two outbreaks of SE phage type 4 likely could have been prevented by using pasteurized eggs and safe food preparation practices.

INTRODUCTION

During the past decade, the number of cases of *Salmonella* serotype Enteritidis (SE) (formerly, *Salmonella enteritidis*) [1] infection has increased substantially in the USA [2]. The proportion of all Salmonella isolates that were SE increased from 9.9% in 1985 to 26.1% in 1994, making SE the most common Salmonella serotype in the USA (CDC, unpublished data). Much of this increase can be attributed to consumption of raw or undercooked eggs [3, 4]. From 1985–93, 83% of SE outbreaks with an identified vehicle were associated with eating eggs [2]. In the United States, the phage types most commonly associated with human outbreaks of SE are phage types 8, 13a, and 13 [4, 5]. Similar increases in SE infections have occurred in the past decade in Europe, where phage type 4 has emerged as the predominant phage type, spreading rapidly through both poultry and human populations and virtually replacing all other phage types [6, 7]. SE phage type 4 has remained rare in the United States [5]. In April 1993, the first recognized US outbreak of SE phage type 4 infections occurred among 130 persons (48 with culture-confirmed infection) who had recently eaten at a fast-food Chinese restaurant in El Paso, Texas. Although food histories were collected from ill persons, a case-control study was not performed, and no source was identified. When a second outbreak of SE phage type 4 infections associated with the same restaurant...
occurred in September 1993, an investigation was performed to determine the source of the outbreaks.

METHODS

Case finding

A case was defined as diarrhoea (three or more loose stools in a 24-h period) or culture-confirmed SE infection that occurred after the patient ate at the restaurant (Restaurant A) between 27 August and 15 September 1993. Cases were identified by means of a press release that asked for persons with a diarrhoeal illness to contact the El Paso City-County Health District and by reviewing all isolates of Salmonella reported since 27 August 1993 to the Health District Laboratory and to the four largest medical laboratories in El Paso.

Case-control study

To determine the source of the outbreak, a case-control study was performed; all identified cases were included, and two sets of controls were used. Control group 1 consisted of 67 persons eating at Restaurant A on 20 or 21 September 1993. Control group 2 consisted of 17 persons who were either well meal companions of case-patients identified during case interviews or persons in control group 1 who recalled eating at the restaurant during the outbreak period, 2–6 September 1993. Using a questionnaire based on the restaurant's menu, we obtained information regarding foods eaten, time of onset of symptoms, and symptom severity. Patients and meal companions were interviewed by telephone between 18 and 27 September 1993. Controls eating at Restaurant A on 20–21 September 1993 were interviewed in person as they left the restaurant, using the same questionnaire.

Laboratory investigations

Salmonella isolates from patients were sent to the Centers for Disease Control and Prevention (CDC) for serotyping [1] and phage typing [8]. Stool samples were collected from all food workers at Restaurant A on 3 consecutive days. These samples, as well as several food items obtained from the restaurant on 7 and 8 September 1993, were cultured at the El Paso Health District Laboratory for salmonella, shigella, and campylobacter. Shell eggs were obtained from the supplier to the restaurant, and 35 batches of 20 eggs each were cultured at CDC.

Restaurant and farm investigations

A restaurant inspection was conducted on 7 September 1993, and a food preparation review was performed on 21 and 24 September 1993. On 6 October 1993 the farm that supplied eggs to the restaurant was inspected according to USDA regulations [9], and 204 environmental samples were collected from manure and egg transport machinery. These were cultured for salmonella at the National Veterinary Services Laboratory in Ames, Iowa [9].

Statistical analysis

Odds ratios (OR) and Cornfield 95% confidence intervals (CI) were calculated using Epi Info version 5 software [10].

RESULTS

Descriptive epidemiology

Nineteen persons met the case definition. All reported having abdominal cramps and 18 (95%) reported having diarrhoea; 5 (26%) had bloody diarrhoea. Other symptoms included headache (90%), myalgias (84%), fever (84%), nausea (79%), and vomiting (42%). The median age of patients was 25 years (range 2-61); 68% were female. Twelve (63%) consulted a physician, and 2 (11%) were hospitalized. The median incubation period was 165 h (range 15-103 h), and the median duration of illness was 4-5 days (range 1-11 days). Figure 1 shows the number of cases by date of onset of illness both for this outbreak and for the one that had occurred in April.

Case-control study results

We analysed the risk of various food items using two control groups (Table 1). The controls were similar in age and sex to the patients. The median age of those in control group 1 was 33 years (range 4-81), and 58% were female; the median age of those in control group 2 was 28 years (range 4-52), and 59% were female. In both analyses, illness was significantly associated with eating egg rolls. In the first analysis, 14 (74%) of 19 case-patients but only 17 (25%) of 67 controls in group 1 ate egg rolls (OR 8.2, CI 2.3–31.6).
Recurrent Salmonella Enteritidis outbreaks

Fig. 1. Cases of gastroenteritis by date of onset – April and September 1993 outbreaks, El Paso, Texas. ■, Culture-confirmed case; □, clinical case.

Breaded chicken was also significantly associated with illness (OR 3.7, CI 1.05–13.5). Neither menu items containing ‘any chicken’ nor items with ‘any breading’ were a significant risk for illness. In the second analysis, 14 (74%) of 19 patients but only 3 (18%) of 17 controls in group 2 ate egg rolls (OR 13.1, CI 2.1–97.0). There was no association between breaded chicken and illness in the second analysis. All seven persons with stool cultures that yielded salmonella reported eating egg rolls.

Data collected on the food eaten by all 48 persons with culture-confirmed infections from the April outbreak were tabulated, and compared with data from control group 1 (Table 2). In this retrospective analysis, egg rolls were the only food associated with illness; 44 (92%) of 48 patients reported eating egg rolls (OR 32.4, CI 9.1–126.6).

Restaurant investigation

Restaurant A is one of a chain of three fast-food Chinese restaurants in El Paso. A sanitation inspection was conducted, and cooks were observed preparing food at Restaurant A as well as at the other two locations (Restaurants B and C). Egg rolls were made by wrapping a premade crepe around refrigerated filling. Using his index finger, the food worker would then place some egg batter in the crease of the crepe to seal it, hand-dip the egg rolls in the batter, and then fry them in hot oil. The batter consisted of six pooled shell eggs, flour, cornstarch, salt, oil, monosodium glutamate, pepper, baking powder, egg shade dye and water. Shell eggs were stored refrigerated at all three restaurants.

Cooks making egg rolls at Restaurant A were observed dipping their hands into the egg batter and then directly into the filling. The filling was stir-fired in a wok and consisted of pork, chicken, sugar, pepper, celery, onions, salt, monosodium glutamate, Chinese spices and cornstarch. Since the filling typically lasts 3–4 days while the batter is made fresh every morning, it is possible that this filling was cross-contaminated during preparation of the egg rolls. In addition, because the same cooks prepared egg rolls and other items, other foods could have been cross-contaminated as well.

At Restaurant A, the egg roll batter was prepared each morning and then kept at room temperature all day. We observed the two cooks who had prepared egg rolls during the outbreak. Frying time ranged from 45–85 seconds, and the internal temperature of egg rolls immediately after cooking ranged from 120–140 °F. At Restaurant A, several egg rolls were made at once and placed in the steam tray at the ordering counter until they were sold. The temperature of the steam holding tray was 110 °F on 7 September 1993.

The procedure for preparing egg rolls at the two other restaurants differed in the following ways: at Restaurant B, the egg roll batter was refrigerated when not in use, the observed frying time was 3 min, and internal temperature of the egg rolls reached 165 °F. At Restaurant C, the egg roll batter was left at room temperature throughout the day; however, the observed cooking time was 2 min 45 seconds, and the internal temperature of the egg rolls was 158 °F.

Laboratory investigation

Of the 19 patients, 7 had stool cultures that yielded Salmonella Enteritidis phage type 4, 4 had negative stool cultures, and 8 had no stool specimen taken. No other cases of Salmonella Enteritidis were identified at the four major medical laboratories in El Paso. All nine food workers working at Restaurant A at the time of the outbreak denied having had a recent diarrhoeal illness, and all had stools collected for culture on 3 consecutive days between 7 September and 16 September 1993; all cultures were negative. All foods obtained from the restaurant on 7 and 8 September 1993 were culture negative, including egg roll batter and chicken used in the filling. No leftover filling was available for testing.

Egg traceback

Because pooling and undercooking shell eggs are known to be risk factors for SE infection [3, 4], eggs
Table 1. Selected menu items at Restaurant A eaten by patients and persons in control groups 1 and 2, El Paso, Texas, September 1993

<table>
<thead>
<tr>
<th>Food item</th>
<th>Patients (n = 19) % exposed</th>
<th>Control group 1 (n = 67) % exposed</th>
<th>OR*</th>
<th>95% CI†</th>
<th>Control group 2 (n = 17) % exposed</th>
<th>OR</th>
<th>95% CI†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Egg rolls</td>
<td>74</td>
<td>74</td>
<td>8.2</td>
<td>2.3-31.6</td>
<td>18</td>
<td>1.5</td>
<td>0.3-8.1</td>
</tr>
<tr>
<td>Breaded chicken</td>
<td>74</td>
<td>43</td>
<td>3.7</td>
<td>1.05-13.5</td>
<td>65</td>
<td>1.5</td>
<td>0.3-8.1</td>
</tr>
<tr>
<td>Any chicken</td>
<td>84</td>
<td>49</td>
<td>1.1</td>
<td>0.2-5.5</td>
<td>94</td>
<td>0.3</td>
<td>0.01-4.4</td>
</tr>
<tr>
<td>Any breaded item</td>
<td>74</td>
<td>49</td>
<td>2.9</td>
<td>0.8-10.6</td>
<td>65</td>
<td>2.1</td>
<td>0.4-11.8</td>
</tr>
<tr>
<td>Fried rice</td>
<td>74</td>
<td>72</td>
<td>1.1</td>
<td>0.3-4.2</td>
<td>53</td>
<td>2.5</td>
<td>0.5-13.0</td>
</tr>
</tbody>
</table>

* OR, odds ratio; † 95% CI, 95% confidence interval.

Table 2. Results of retrospective case-control study; selected menu items at Restaurant A eaten by patients with culture-confirmed infection and persons in control group 1, El Paso, Texas, April 1993

<table>
<thead>
<tr>
<th>Food item</th>
<th>Patients (n = 48) % exposed</th>
<th>Control group 1 (n = 67) % exposed</th>
<th>OR*</th>
<th>95% CI†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Egg rolls</td>
<td>92</td>
<td>25</td>
<td>32.4</td>
<td>9.1-126.6</td>
</tr>
<tr>
<td>Breaded chicken</td>
<td>44</td>
<td>43</td>
<td>1.0</td>
<td>0.5-2.3</td>
</tr>
<tr>
<td>Any chicken</td>
<td>69</td>
<td>84</td>
<td>0.4</td>
<td>0.2-1.2</td>
</tr>
<tr>
<td>Any breaded item</td>
<td>44</td>
<td>49</td>
<td>0.8</td>
<td>0.4-1.8</td>
</tr>
<tr>
<td>Fried rice</td>
<td>56</td>
<td>72</td>
<td>0.5</td>
<td>0.2-1.2</td>
</tr>
</tbody>
</table>

* OR, odds ratio; † 95% CI, 95% confidence interval.

DISCUSSION

In April and September 1993, two outbreaks of Salmonella Enteritidis phage type 4 infection occurred among patrons of Restaurant A in El Paso, Texas. Although sporadic cases of infection have been reported in the USA, these are the first two SE phage type 4 outbreaks reported in this country that were not associated with foreign travel [11, 12]. For the September outbreak, egg rolls were the only item associated with illness in both case-control studies, and egg rolls were also retrospectively implicated in the April outbreak. Inspection of the procedure for egg roll preparation strongly suggests that egg batter made at Restaurant A was the source of the September outbreak. Of the ingredients used to make the batter, shell eggs are the most likely source of salmonella contamination because the other ingredients have not been associated with SE infections, whereas eggs have been commonly associated with SE infections throughout the USA [2-4, 13-16]. Eggs have also been
associated with sporadic and outbreak-associated cases of SE in Europe [6, 17].

Several poor food preparation techniques may have contributed to the occurrence of outbreaks at Restaurant A but not at Restaurants B or C. First, batter at Restaurant A was left at room temperature throughout the day. Second, cooking times and temperatures were inadequate to kill salmonella [18]. In comparison, Restaurant B refrigerated the egg batter and adequately cooked the egg rolls. Although Restaurant C did not refrigerate the batter, the cooking time and temperature were sufficient to kill salmonella. Finally, inadequate holding temperatures in the steam trays in Restaurant A may have allowed salmonella organisms that survived cooking to increase in number.

Another poor food preparation technique observed at Restaurant A (dipping hands into the egg batter and then directly into the filling) may explain how cross-contamination enabled the outbreak to continue for several days, even though the egg batter was made fresh every morning. Cross-contamination may also have been responsible for breaded chicken being weakly associated with illness in one of the case-control studies. SE phage type 4 has been recovered from fingers, utensils, foods and work surfaces up to 24 h after contamination [19], illustrating the potential for cross-contamination.

These lapses in proper food preparation techniques are instructive. Despite contamination of eggs with SE, outbreaks due to this organism are preventable by adhering to safe food preparation practices, such as avoiding the pooling of eggs. Pooling eggs enables a large number of persons to be infected from a single contaminated egg; restaurants should not use pooled shell eggs but should use pasteurized egg product instead. No reported cases of SE infection have been traced to the use of pasteurized eggs. In 1990, the US Food and Drug Administration Model Food Code redesignated shell eggs as a potentially hazardous food and recommended that food establishments be required to keep eggs refrigerated [20]. To prevent egg-associated SE outbreaks from occurring, food workers must be instructed in proper handling of eggs; this includes avoidance of using pooled eggs, proper refrigeration, avoidance of cross-contamination with other foods, thorough cooking and adequate holding temperatures.

In England and Wales, the number of reported Salmonella Enteritidis isolates has increased 16-fold, from 1101 in 1982 to 17369 in 1994 [7, 21]. SE phage type 4 accounted for most of this increase; previously rare, SE phage type 4 now accounts for 79% of SE isolates reported to the Communicable Disease Surveillance Centre [7]. The reason for this epidemic of SE phage type 4 is unclear; some have postulated that recent strains of SE phage type 4 are more invasive for young chickens, facilitating transovarial transmission to the next generation [22, 23]. Whatever the mechanism, the current epidemic in European countries suggests that once SE phage type 4 is introduced into a population of layer chickens, it has the ability to spread rapidly, infecting both poultry and human populations, and to persist as the dominant strain in those populations. To detect the beginnings of a similar epidemic in the USA, we encourage the prompt reporting of SE outbreaks and the forwarding of selected isolates to CDC for phage typing.

In 1990, USDA began an SE control programme based on tracing eggs implicated in human outbreaks to source egg farms, testing the farm environment and the internal organs of chickens on those farms, and diverting the eggs to pasteurization if cultures yield SE [9]. Since the implementation of this programme, it has been unusual for a farm that has been epidemiologically linked to an SE outbreak to test negative for SE. In a retrospective study of 18 SE outbreaks epidemiologically associated with shell eggs in 1990–1, the predominant human outbreak phage type was recovered from the environment in 100%, and from the internal organs of hens in 88% of implicated flocks tested [5]. That the traceback for this outbreak was negative raises the possibility (although denied by the restaurant’s owners) that eggs from Mexico, which borders El Paso, were the source of the salmonella in this outbreak. Because phage typing is not routinely performed in Mexico, the prevalence of phage type 4 in that country is unknown, but it is thought to be greater than in the USA. In 1990, a convenience sample of SE isolated from persons in Mexico showed that 22 (79%) of 28 isolates were phage type 4 (CDC, unpublished data).

In summary, two outbreaks of Salmonella Enteritidis phage type 4 infection were caused by egg rolls eaten at a Chinese restaurant, and pooled shell eggs were the most likely source of contamination. These outbreaks likely could have been prevented by using pasteurized eggs and safe food preparation practices. As shown by the current epidemic in Europe, the introduction of SE phage type 4 into US breeder flocks could have a dramatic effect on the epidemiology of salmonella infections, which emphasizes...
the need for continued surveillance for the spread of this pathogen in the USA, both in humans and in the layer industry.

ACKNOWLEDGEMENTS

We are indebted to Due Vugia of the California State Department of Health; Suzanne Barth of the Texas Department of Health Laboratory; Jeffery Taylor, Kate Hendricks, and Diane Simpson of the Texas Department of Health Laboratory; Jeffery We are indebted to Due Vugia of the California State

REFERENCES


