

FOURTH EPIDEMIOLOGICAL PROBLEM SET

Problem	Title	Page
13	GASTROENTERITIS ASSOCIATED WITH EATING OYSTERS — LOUISIANA, DECEMBER 1996-JANUARY 1997	2
14	FOODBORNE OUTBREAK OF DIARRHEAL ILLNESS — MINNESOTA, 1995	5
15	NEW OUTBREAK IN THE MIDI-PYRÉNÉES REGION OF FRANCE, SEPTEMBER - OCTOBER 1998	7
16	OUTBREAKS OF INFECTION ASSOCIATED WITH EATING FRESH PARSLEY — UNITED STATES AND CANADA, JULY-AUGUST 1998	10

**GASTROENTERITIS ASSOCIATED WITH EATING OYSTERS — LOUISIANA,
DECEMBER 1996-JANUARY 1997**

Gastroenteritis outbreaks have been associated with eating contaminated shellfish, particularly oysters (*Crassostrea virginica*). This report describes the findings of the investigation of an outbreak of oyster-associated gastroenteritis in Louisiana during the 1996-97 winter season and implicates sewage from oyster harvesting vessels as the probable cause of contaminated oysters.

On December 30, 1996, the Louisiana Office of Public Health (LOPH) was notified about a cluster of six persons who had onset of gastroenteritis after eating raw oysters on December 25. During December 30, 1996-January 3, 1997, three additional clusters were identified. In all four clusters, ill persons had eaten oysters harvested from Louisiana waterways. LOPH notified all state epidemiologists in the United States about the apparent association of gastroenteritis with eating oysters and requested reports of suspected cases.

A case of gastroenteritis was defined as three or more watery stools or vomiting within a 24-hour period, with onset during December 15-January 9. A cluster of oyster-related cases was defined as a group of three or more persons who had shared a common meal, at least one of whom had eaten oysters and at least one of whom developed gastroenteritis. Sixty clusters comprising 493 persons were reported from Alabama, Florida, Georgia, Louisiana, and Mississippi, and all were included in the subsequent traceback investigation. Of the 60 clusters, data were included in the descriptive analysis of the illness only for those 34 clusters for whom all persons in a cluster could be interviewed. The 34 clusters comprised 290 persons who completed interviews and were included in the descriptive analysis; 271 of 290 persons supplied information on oyster consumption.

Onsets of illness occurred during December 21-January 7 (Figure 1). Of the 290 persons interviewed, 179 (62%) had symptoms that met the case definition. The most common symptoms were diarrhea (83%), abdominal cramps (78%), vomiting (58%), headache (50%), and fever (50%). The median incubation period was 38 hours (range: 8-90 hours), and the median duration of illness was 2 days (range: 1-14 days). The median age of case-patients was 42 years (range: 14-83 years), and 111 (62%) were male. The number of reported cases peaked during December 31-January 5 (Figure 1); the harvest dates of subsequently implicated oysters ranged from December 15 to January 1. Of 201 persons who ate raw oysters, 153 (76%) became ill, compared with 13 (19%) of 70 persons who did not eat raw oysters (risk ratio=4.0). Small round-structured viruses were found by direct electron microscopy in fecal specimens from eight of 11 ill persons. Sequence analysis of nucleic acid from eight specimens representing six clusters demonstrated three unique genetic sequences that corresponded with oysters harvested from three separate harvest sites. Agent F was detected in oysters, but genetic sequencing could not be conducted.

The LOPH traced oysters eaten by ill persons to retailers, wholesalers, and harvesters. Restaurants and seafood markets were inspected to observe handling and storage of shellfish, and tags that identified the date and site of harvesting and the harvester's identification number were obtained from purchasers and retailers of sacks that were definitely or possibly implicated. Retailer records were cross-referenced with records from wholesalers and harvesters to establish the accuracy of information about harvester and site of harvest. Oysters associated with the 60 clusters were traced to 26 retailers, 11 wholesalers, and 20 harvesters. Records from several wholesalers did not agree with the information on the oyster sack tag.

As of February 15 (6 weeks after notification of the outbreak), LOPH, despite repeated attempts, had been successful in completing interviews with only three of 20 harvesters about the date and specific location of harvesting of potentially contaminated oysters. However, with the assistance of Louisiana Department of Wildlife and Fisheries, 12 additional harvesters were interviewed. Of eight oyster harvesting boats inspected, seven had inadequate sewage collection and disposal systems.

Testing by the LOPH Molluscan Shellfish Program determined that a toxic algal bloom, which causes paralytic shellfish poisoning, was present in Louisiana's northeastern waterways beginning November 13, 1996; these findings prompted LOPH to close these waterways that day and required harvesters to move to southeastern harvest sites. In addition, on November 15, a freshwater diversion was opened to decrease the salinity and eliminate the algal bloom in the northeastern waters; the diversion also decreased the salinity in the southeastern waters.

On January 3, 1997, LOPH mandated an emergency closure of eight waterways with suspected contamination southeast of the Mississippi River, and on January 6, LOPH recalled oysters harvested from these sites after December 22, 1996. On January 23, 1997, harvesting was permitted to resume, and no additional cases of oyster-associated gastroenteritis were reported.

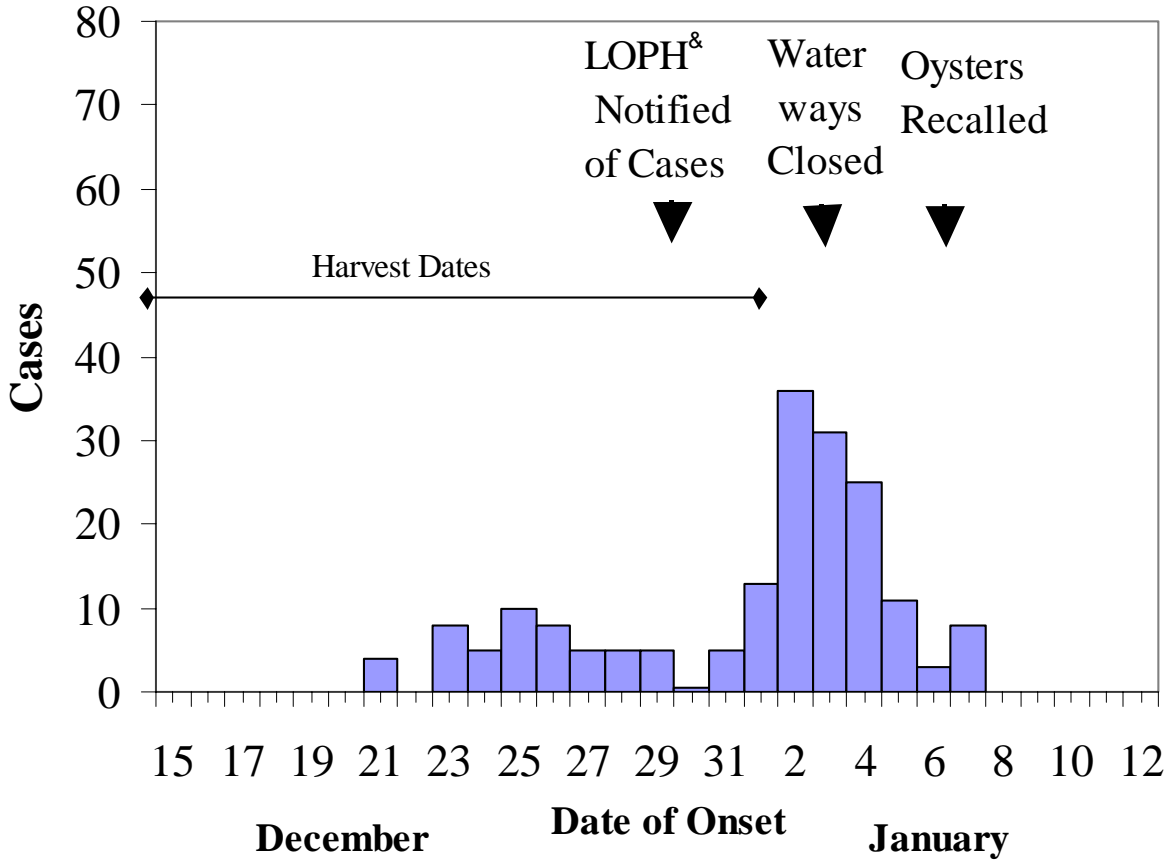
1. What was the probable Agent F of these illnesses?

2. Oyster harvesting is prohibited near sewage outfalls. Why did that not protect consumers in this instance?

3. What do you suppose is meant by “inadequate sewage collection and disposal systems,” and what could the authorities do about this?

4. Could the shellfish be tested for contamination?

Figure 1. Number of cases of gastroenteritis* associated with eating oysters harvested from Louisiana waterways, December 1996-January 1997^t



FOODBORNE OUTBREAK OF DIARRHEAL ILLNESS — MINNESOTA, 1995

On September 29, 1995, the Minnesota Department of Health (MDH) received reports of acute gastroenteritis among an estimated 50 attendees of a social event in Blue Earth County on September 16. This report summarizes the epidemiologic and laboratory investigations of the outbreak.

Of the 26 persons who attended the function and who completed telephone interviews with MDH, 15 (58%) reported onset of diarrhea (three or more stools during a 24-hour period) within 14 days after attending the event (range: 1-9 days; median: 6 days). Symptoms included watery diarrhea (100%), abdominal cramps (93%), and chills (79%). The median length of illness was 4 days (range: 1/2 day-14 days). Three persons who sought medical care received outpatient treatment for acute gastroenteritis. Stool specimens obtained from two of these persons were negative for bacterial pathogens and for ova and helminth parasites.

To identify risk factors for illness, MDH conducted a case-control study using the 15 ill and 11 well attendees. In addition, MDH collected stools from three ill persons, and these were cultured for *Salmonella*, *Shigella*, *Campylobacter*, and *Escherichia coli* O157:H7; examined for ova and parasites; and tested for “agent Q” using DFA methods.

Based on the case-control study, only consumption of chicken salad was associated with increased risk for illness (15 of 15 cases versus two of 11 controls; odds ratio= undefined).

Water consumption at the event was not associated with illness.

The chicken salad was prepared by the hostess on September 15 and was refrigerated until served. The ingredients were cooked chopped chicken, pasta, peeled and chopped hard-boiled eggs, chopped celery, and chopped grapes in a seasoned mayonnaise dressing. The hostess operated a licensed day-care home (DCH) and prepared the salad while attendees were in her home. She denied having recent diarrheal illness and refused to submit a stool specimen. In addition, she denied knowledge of diarrheal illnesses among children in her DCH during the week before preparation of the salad. She reported changing diapers on September 15 before preparing the salad and reported routinely following handwashing practices.

Stool specimens from two of the persons whose illnesses met the case definition were obtained by MDH 7 days after resolution of their symptoms; one sample was positive for agent Q. Stool specimens obtained from a third person — the spouse of a case-patient — who did not attend the event but had onset of diarrhea 8 days after onset of diarrhea in his spouse was positive for agent Q. All stools obtained by MDH were negative for bacteria and for parasites. No chicken salad was available for testing.

1. What was the probable etiologic agent of this outbreak?

2. Do you think the source of contamination of the chicken salad was most likely one of the ingredients, the woman who prepared it, or the “DCH attendees”?

3. Might inadequate refrigeration of the salad before serving have played a role in this outbreak?

**NEW OUTBREAK IN THE MIDI-PYRÉNÉES REGION
OF FRANCE, SEPTEMBER - OCTOBER 1998**

Eleven foodborne illness cases in the Haute-Garonne, and Tarn districts of France were reported to the regional health department of Midi-Pyrénées region on 6 October. All cases had eaten horse meat, bought in one butcher's in Toulouse and one in Castres. An epidemiological, veterinary and parasitological investigation was conducted to assess the outbreak's importance and identify the vehicle and source of infection.

Cases were defined as residents of Tarn and Haute-Garonne districts who had presented with the following features since 1 September 1998 :

- Confirmed case: fever ($>38^{\circ}$) with myalgia or facial oedema.
- Probable case: at least three out of the following four criteria : fever ($>38^{\circ}$), myalgia, facial oedema, hypereosinophilia $>1000/\text{mm}^3$.
- Suspected case: hypereosinophilia $>1000/\text{mm}^3$ alone or associated with fever or myalgia.

Cases were sought actively by the Cellule Interrégionale d'Epidémiologie d'Intervention du Sud-Ouest (CIREI) among medical laboratories, general practitioners and hospital physicians in the Haute-Garonne and Tarn districts. They were asked to report hypereosinophilia ($>1000/\text{mm}^3$) and patients who had consulted for symptoms. Hospital pharmacists were also asked to report prescriptions for albendazole.

A standardized questionnaire was administered by telephone to the cases identified who could be contacted at the time of the study. Subjects were asked about clinical features, dates of onset of symptoms, laboratory tests performed, consumption of meat products, and where and when they had bought meat in September.

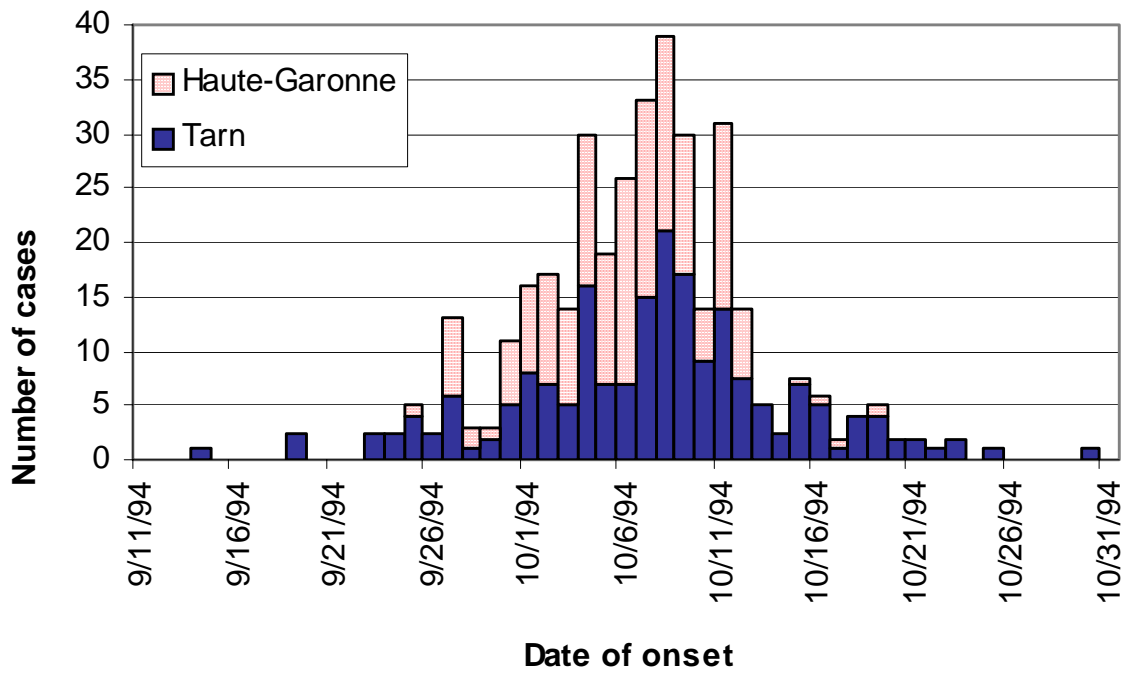
The departmental veterinary services took samples from meat bought by cases in September and kept in their freezers. The distribution channels were identified from the purchasing sites reported by cases. Biopsies were taken from pets of two cases, a cat and a dog, that had been fed horse meat. The samples were tested using the enzymatic digestion method.

Four hundred and four cases were identified who lived in the two districts of the Midi-Pyrénées region and had become ill between 20 September and 27 October. Thirty seven cases were admitted to hospital and one case suffered neurological complications. The epidemic curve suggested the contamination had a point source in time in the third week of September (figure). All cases had eaten horse meat steaks bought by cases. Analysis performed by the Centre National d'Etudes Vétérinaires et Alimentaires showed a high level of contamination (900 to 2700 larvae per 100g of horse meat). Biopsy from pets both yielded larvae – one larva in 0.1 g from the dog; 11 larvae in 0.4 g from the cat. Investigation of the supply and distribution

channels identified a horse carcass from a batch of horses imported from the Federal Republic of Yugoslavia and slaughtered in France. A previous outbreak, in the same region of France in February 1998, was linked to the consumption of horse meat imported from the same country.

1. What was the pathogen?
2. Do you agree with: “The epidemic curve suggested the contamination had a point source in time in the third week of September”? Why or why not?
3. What was the purpose of asking hospital pharmacists to report prescriptions for albendazole?
4. How would a horse become infected with the pathogen?
5. Other than not eating horsemeat, what could have been done to prevent these illnesses?

Figure 1.
Epidemic curve by onset of symptoms,
Midi-Pyrénées, France, September-October 1998.



**OUTBREAKS OF INFECTION ASSOCIATED WITH EATING FRESH PARSLEY —
UNITED STATES AND CANADA, JULY-AUGUST 1998**

In August 1998, the Minnesota Department of Health reported to CDC two restaurant-associated outbreaks. Isolates from both outbreaks had two closely related pulsed-field gel electrophoresis (PFGE) patterns that differed only by a single band. Epidemiologic investigations implicated chopped, uncooked, curly parsley as the common vehicle for these outbreaks. Through inquiries to health departments and public health laboratories, six similar outbreaks were identified during July-August (in California [two], Massachusetts, and Florida in the United States and in Ontario and Alberta in Canada). Isolates from five of these outbreaks had the same PFGE pattern identified in the two outbreaks in Minnesota. This report describes the epidemiologic, traceback, environmental, and laboratory investigations, which implicated parsley imported from a farm in Mexico as the source of these outbreaks. Outbreak investigations showed that, overall, the symptoms of ill individuals included diarrhea, fever, toxemia, vomiting, cramps, and tenesmus.

United States

Minnesota. On August 17, the Minnesota Department of Health received reports of foodborne illness in two persons who ate at the same restaurant during July 24-August 17. A pathogen subsequently was isolated from stool samples of 43 ill restaurant patrons; an additional 167 persons had probable foodborne illness (diarrhea [three or more loose stools during a 24-hour period] lasting ≥ 3 days or accompanied by fever). Eight (18%) of 44 restaurant employees had a similar illness; five had laboratory-confirmed pathogen infection. In a case-control study of 172 ill and 95 well restaurant patrons, five items were associated with illness: water (odds ratio [OR]=1.9; 95% confidence interval [CI]=1.0-3.8), ice (OR=3.7; 95% CI=1.6-8.6), potatoes (OR=2.6; 95% CI=1.5-4.6), uncooked parsley (OR=4.3; 95% CI=2.4-8.0), and raw tomato (OR=1.9; 95% CI=1.0-3.9). In a multivariate analysis, only uncooked parsley (OR=4.3; $p < 0.01$) and ice (OR=6.9; $p < 0.01$) remained significantly associated with illness.

California. On August 5, the Los Angeles County Department of Health Services was notified of two persons with foodborne illness who ate at the same restaurant on July 31. Stool samples from six ill restaurant patrons yielded a foodborne pathogen; an additional three had probable illness (diarrhea [three or more loose stools during a 24-hour period], or any loose stools accompanied by fever). All 27 food handlers denied illness and had stool samples that were negative for the pathogen. In an unmatched comparison with 10 well dining companions, ill patrons were significantly more likely to have eaten foods sprinkled with chopped, uncooked parsley (OR=32.0; 95% CI=1.8-1381.4).

Massachusetts. On August 11, the Massachusetts Department of Health was notified of six

persons who reported illness after eating at a restaurant lunch party on July 30. Stool samples from three persons yielded a foodborne pathogen; an additional three had diarrhea within 4 days of the July 30 meal. Chopped, uncooked parsley was served on chicken sandwiches and in cole slaw served at the lunch. In a cohort study of 23 lunch attendees, illness was significantly associated with eating chicken sandwiches (relative risk [RR]=10.0; 95% CI=2.7-37.2) or eating uncooked parsley with any item (RR=10.0; 95% CI=1.4-70.2). All restaurant employees except one submitted a stool sample for culture; all were negative for the pathogen.

Canada

On August 10, the Ontario Ministry of Health was notified of a family of three persons with foodborne infection who attended a food fair during July 31-August 3. Laboratory-based surveillance identified 32 additional persons with infection who had eaten at a specific kiosk at the fair or at the restaurant that had supplied the kiosk. Of the 35 persons, 20 were questioned about food history; all reported eating a smoked salmon and pasta dish made with fresh chopped parsley. Stool samples from six (38%) of 16 food handlers, including the four who handled the parsley, were negative for the pathogen. One child who had eaten at the kiosk was the index patient at a day care center, from which five secondary cases of foodborne illness were reported.

Other Investigations

In addition to these four outbreaks, four additional restaurant-associated outbreaks of the same pathogen were identified, involving an additional 218 persons with culture-confirmed or probable illness. Of the 111 persons interviewed, 106 (96%) reported eating chopped, uncooked, curly parsley. Isolates from three of these outbreaks (in Minnesota and California in the United States and in Alberta in Canada) matched the outbreak PFGE pattern. In the fourth outbreak (in Florida), one culture-confirmed case was identified; the isolate was not available for PFGE testing.

Traceback and Environmental Investigations

To determine the source(s) of parsley for the seven outbreaks linked by PFGE, state and provincial health departments, CDC, the Food and Drug Administration (FDA), and the Canadian Food Inspection Agency conducted traceback investigations. Farm A in Baja California, Mexico, was a possible source of parsley served in six of the seven outbreaks; four farms in California were possible sources of parsley in two to four of the seven outbreaks.

Field investigations of farm A by FDA and CDC found that the municipal water that supplied the packing shed was unchlorinated and vulnerable to contamination. This water was used for chilling the parsley in a hydrocooler immediately after harvest and for making ice with which the parsley was packaged for transport. Because the water in the hydrocooler was recirculated, bacterial contaminants in the water supply or on the parsley could have survived in the absence of chlorine and contaminated many boxes of parsley. Farm workers and village residents served by this water system reported drinking bottled water or water from other

sources. Workers had limited hygiene education and limited sanitary facilities available on the farm at the time of the outbreak.

Food handlers at six (75%) of the eight implicated restaurants reported washing parsley before chopping it. Usually parsley was chopped in the morning and left at room temperature, sometimes until the end of the day, before it was served to customers.

Laboratory Investigations

The Minnesota Department of Health laboratory, which has tested isolates of the pathogen by PFGE routinely since 1995, identified a previously unrecognized PFGE pattern of the pathogen and a closely related pattern that differed by a single band associated with the two outbreaks in Minnesota. The pattern was distributed to other laboratories through PulseNet, the national molecular subtyping network for foodborne disease. In Minnesota and at CDC, strains from all seven outbreaks for which isolates were available for PFGE testing had the outbreak PFGE pattern.

Investigators at the University of Georgia Center for Food Safety and Quality Enhancement conducted studies to determine the effects of temperature and handling on the growth and survival of the pathogen on parsley. Colony-forming units of the pathogen per gram (CFU/g) decreased by approximately 1 log per week on parsley, whether chopped or whole, under refrigeration (39°F [4°C]). In contrast, counts increased on parsley kept at room temperature (70°F [21°C]). On whole parsley, the increase was limited to 1 log CFU/g during the first 1-2 days, but on chopped parsley a 3 log CFU/g increase was observed within 24 hours.

1. What was the pathogen?
2. How many victims were recorded in each of the involved geographic areas?
3. Does the case definition agree with the description in your text?
4. How many food handlers were tested, and how many were probably positive?
5. How many samples of parsley yielded the pathogen?
6. What do you suppose a “hydrocooler” is, and how might the water have gotten contaminated with this pathogen?
7. Should hydrocoolers be used in restaurants?