An Outbreak of *Escherichia coli* O157:H7 Infection Linked to Unpasteurized Apple Cider in Oklahoma, 1999

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**ABSTRACT**

During the fall of 1999, an outbreak with 16 cases (11 confirmed and five probable) of *Escherichia coli* O157:H7 infections was identified in Oklahoma. Nine persons (82%) experienced bloody diarrhea, six (54%) required hospitalization, and three (27%) developed hemolytic uremic syndrome (HUS). Twelve of the sixteen (75%) cases were children from 2 to 13 years old. All nine available *E. coli* O157:H7 isolates had an identical pulsed-field gel electrophoresis pattern. Ten of the 11 confirmed patients and none of the 24 control subjects had drunk unpasteurized apple cider from Orchard A (matched odds ratio undefined; \( P < 0.00001 \)). All environmental specimen cultures were negative. Orchard A was in compliance with U.S. Food and Drug Administration requirements for product warning labels on unpasteurized products, although only one of eight patients (or surrogates) interviewed recalled having read the label. This outbreak raises questions about whether the current practice of requiring warning labels for unpasteurized cider provides sufficient protection for vulnerable populations, especially children.

**INTRODUCTION**

In 1982, Riley et al. (22) investigated two outbreaks of an unusual gastrointestinal illness that affected 47 or more persons. They determined that the outbreak was caused by a rare *E. coli* serotype, O157:H7, apparently transmitted by undercooked meat. During the following decades, multiple *E. coli* O157:H7 outbreaks were associated with consumption of raw or undercooked ground meat, meat products, and unpasteurized milk (2, 3, 15, 16, 17). After the 1990s, an increased number of reported outbreaks were associated with fresh vegetables (e.g., lettuce, spinach, sprouts) or unpasteurized apple juice (1, 4, 5, 6, 8, 11, 12, 13, 19, 24).

In 1999, Mead et al. (18), from the Centers for Disease Control and Prevention (CDC), estimated that *E. coli* O157:H7 accounted for 73,480 illnesses annually in the United States, including 2,000 hospitalizations and 60 deaths, as a direct result of *E. coli* O157:H7 infections and its complications. Despite progress in making food safer in the United States, *E. coli* O157:H7 associated with animal products and fresh produce remains a public health challenge putting a large number of persons at risk.
for *E. coli* O157:H7 infection and its complications (7, 11, 14, 21).

In Oklahoma, from 1994 through 1998, an average of 17 cases of *E. coli* O157:H7 infections were reported to the Oklahoma State Department of Health (OSDH) annually. On October 11, 1999, a hospital laboratory in northeast Oklahoma notified OSDH of four patient specimens that had tested positive for *E. coli* O157:H7 by a fecal antigen-screening assay. In response to these reports, OSDH immediately initiated an epidemiologic investigation to (1) determine if these represented an outbreak and if so, the magnitude of the outbreak, (2) identify risk factors for illness and the possible source or vehicle for infection, and (3) implement preventive measures.

**MATERIALS AND METHODS**

**Case finding**

After the initial case report, OSDH initiated active surveillance. All hospitals and clinical laboratories in Tulsa and neighboring counties in Oklahoma were contacted by telephone and encouraged to report suspected cases of *E. coli* infection or postdiarrheal hemolytic uremic syndrome (HUS). A confirmed case was defined as diarrheal illness (more than three loose stools per 24 hours) occurring among persons of Northeastern Oklahoma (Tulsa and neighboring counties) during October 1999, and a positive fecal antigen test or culture for *E. coli* O157:H7. A probable case was defined as diarrhea (three or more loose stools per 24 hours) occurring in a family member of a patient with confirmed illness who was ill at approximately the same time as that patient, but without laboratory confirmation.

**Case-control study**

OSDH conducted a matched case-control study to determine possible risk factors and sources for acquiring infection with *E. coli* O157:H7. For each case-patient, two to three acquaintance control subjects were selected and matched by age and neighborhood. The control subjects’ names and contact information were provided by the index case-patient. All case-patients and control subjects were asked questions regarding clinical symptoms and risk factors for illness, including animal contact, food exposures, and recreational water exposure. All control subjects were asymptomatic and without diarrhea history from October 1 to the date of interview.

During October 12–25, a total of 16 case-patients (11 confirmed and five probable) and 24 control subjects were interviewed by use of a standard questionnaire. If the age of the case-patient or control subject was less than 14 years, a parent/guardian responded to the questions. Case-patients were asked questions regarding food consumption within 7 days of illness onset. Control subjects were asked to report on foods consumed during the 7-day period before the interview.

**Environmental investigation**

After epidemiologically linking the outbreak to consumption of unpasteurized apple cider from a single orchard (defined here as Orchard A), staff from the OSDH Consumer Protection Division, the Wagoner County Health Department, and the OSDH Communicable Disease Division conducted separate site visits and inspections of the suspected orchard. Unopened containers of cider, environmental swabs of the cider production equipment, and samples of tap water from the cider mill were collected for laboratory analyses.

**Laboratory investigation**

All available patients’ stools were cultured at the county laboratory for diarrheal pathogen identification. Testing included direct culture attempts, Shiga-toxin testing (ImmunoCard STAT™ EHEC, Meridian Diagnostics, Inc., Cincinnati, Ohio), and *E. coli* O157:H7 isolation attempts by using immunomagnetic beads provided by the Foodborne Diseases Laboratory, CDC. Positive *E. coli* O157:H7 tests were referred to the OSDH Public Health Laboratory for confirmation. Isolates cultured from available case-patient stools and confirmed as *E. coli* O157:H7 by testing at the OSDH laboratory were forwarded to the Texas Department of Health Laboratory for pulsed-field gel electrophoresis (PFGE) subtyping. PFGE subtypes were compared among outbreak cases and with previously identified cases that had occurred earlier in the year. All XbaI patterns were forwarded to the PFGE *E. coli* national database at the national network of public health and food regulatory agency laboratories coordinated by CDC (PulseNet).

The Oklahoma Dairy, Food, and Water Laboratory and the OSDH Public Health Laboratory tested the environmental swabs and samples of the apple cider collected during the environmental investigation. They performed total and fecal coliform testing and testing for *E. coli* O157:H7 antigen, using the visual immunoprecipitation assay (VIP™: BioControl System, Bellevue, WA, USA) and direct plating of samples from eight unopened containers of apple cider (multiple lot numbers) and one cider sample collected from a case-patient’s household. Bacterial culture and antigen testing were also performed on 20 environmental swabs collected during the investigation (unopened juggs of cider, environmental swabs of cider production equipment, and samples of tap water from the cider mill). Additionally, coliform counts and bacterial culture were performed on tap-water samples collected from the cider processing room and packing shed.

**Statistical analysis**

Data were entered into Epi-Info™ 6.04b (CDC, Atlanta, Georgia). A descriptive analysis was performed to calculate frequency of selected variables. Only confirmed cases were included in the matched case-control analysis of possible risk factors to determine matched odds ratios (ORs) and their 95% confidence intervals (95% CIs).

**RESULTS**

**Active surveillance**

Sixteen cases of *E. coli* O157:H7 infection (11 confirmed and five probable) were identified during October 12–25, 1999, with diarrhea onsets during October 2–17. Reported clinical symptoms of the 11 confirmed cases included abdominal cramping (91%), bloody diarrhea (82%), fever (73%), and vomiting (55%). Duration of diarrheal illness ranged from 2 days to 13 days (mean = 5 days). Six persons (55%) were hospitalized, including three children with hemolytic uremic syndrome requiring dialysis treatment. Twelve (75%) were children from 2 to 13 years old. No other vulnerable population was identified during this outbreak.

**Case-control study**

All 11 confirmed case-patients and 24 control subjects were included in the
case-control study. Case-patients and control subjects had similar sociodemographic characteristics. All the case-patients and controls were residents of two counties in northern Oklahoma. The median age was 10 years, with a range of 2–38 years, for the cases vs. 9 years, with a range of 2–42 years for controls. The majority of participants were female; (64% of the cases vs. 62% for controls) and all were white. Ten (91%) of the 11 case-patients reported drinking apple cider within one week before onset of diarrheal illness, and only two (8%) of 24 control subjects reported drinking apple cider before the interview. All case-patients with this exposure identified the same orchard as the source of the apple cider they had consumed. The two control subjects who had consumed apple cider described having obtained their apple cider from a different orchard.

Analysis of possible risk factors revealed that consumption of apple cider produced at a single orchard (defined here as Orchard A) was discovered to be strongly associated with E. coli O157:H7 infection (matched OR undefined; unmatched P-value < 0.00001). No association with other factors was identified.

On the basis of this epidemiologic evidence, OSDH ordered Orchard A to discontinue unpasteurized apple cider production and recalled the apple cider on October 12, 1999. A press release was issued advising persons not to drink purchased unpasteurized apple cider from local grocery outlets in the outbreak geographic area. The last case of E. coli O157:H7 illness was reported on October 17, 1999, a total of 5 days after Orchard A's unpasteurized apple cider recall.

Environmental investigation

During the environmental investigation, the owner reported that Orchard A, a family-owned farm, had been producing apple cider for approximately 20 years before this outbreak. Orchard A comprised approximately 15–20 acres. Cattle were pastured approximately 200 yards from the orchard. No evidence (tracks or manure) existed to indicate recent presence of deer or cattle in the orchard area. Rain water flowed out of the orchard and toward a pasture adjacent to the orchard. Before the outbreak, five employees had worked on the farm and reported working there during the last 10 years. No new staff were recently hired; none reported recent history of gastrointestinal illness or diarrhea, travel, absenteeism, or contact with animals.

Employees described using public toilet facilities with hand-washing sinks on the farm.

The owner reported that no apples were obtained from other orchards, and no apples retrieved from the ground (known as droppers) were used for cider production before the outbreak. Employees had picked apples from trees, placed them in wooden crates, and sent them to the apple washer facility, which used municipal running water. After washing, the apples were transported in pickup trucks to the cider press approximately 50 meters away.

During the cider production process, potassium polysorbate was added as a preservative, but the juice was unpasteurized. Then the cider was stored in a refrigerated main container of about 270 gallons. At the end of the 1-day process, the juice was dispensed from the main container to half- or one-gallon plastic containers identified by using a Julian system of dating. All containers of unpasteurized cider were labeled with the standard Food and Drug Administration (FDA)-required warning label for persons at highest risk for experiencing foodborne illnesses (i.e., “Warning: This product has not been pasteurized and, therefore, may contain harmful bacteria that can cause serious illness in children, the elderly, and persons with weakened immune systems.”) (25).

Detailed written distribution records were not produced, but according to verbal reports, distribution of the cider was limited to retail grocery outlets.

After identification of Orchard A unpasteurized apple cider as the possible vehicle of contamination, eight study participants and/or their parents/guardians were contacted and asked if they had read the warning label on the apple cider container. Only one out of the eight reported having read the warning label, and then only after a portion of the cider had already been consumed.

Laboratory analyses

Nine stools (82%) out of the 11 stool specimens collected from confirmed cases had positive cultures for E. coli O157:H7, seven (64%) had positive fecal E. coli O157:H7 antigen tests and five (45%) had both antigen test and culture positive. PFGE testing performed on nine available isolates of E. coli O157:H7 cultured from case-patients' stool specimens exhibited Xba-I patterns that were indistinguishable from each other. This was classified as the outbreak strain. Four E. coli O157:H7 isolates from sporadic cases reported during the outbreak period from other regions of the state were also subtyped by PFGE. One of these four displayed a pattern consistent with the outbreak strain. Testing of all apple cider and environmental samples collected during the environmental investigation of Orchard A failed to detect E. coli O157.

DISCUSSION

This investigation confirmed that a regional E. coli O157:H7 outbreak occurred in Oklahoma during October 1999. Sixteen cases (11 confirmed and 5 probable) were identified as outbreak-associated. The investigation provided epidemiologic evidence that unpasteurized apple cider produced and bottled at a local family orchard and then sold to local grocery outlets was the most likely vehicle of the outbreak. Results of the PFGE subtyping on patients' isolates were compatible with a point-source outbreak and consistent with the epidemiologic findings. Only one confirmed adult case-patient, whose diarrhea began October 15, 1999, did not report apple cider consumption; this case-patient lived in the same zip code area as two other confirmed case-patients, whose diarrhea had started October 8 and 9, respectively. We did not investigate whether contact occurred among these three patients. In prior reported E. coli O157:H7 outbreaks (20, 23), secondary cases have been identified, and the case-patient without apple cider consumption might have been a secondary case.

Similar outbreaks of E. coli O157:H7 infection associated with consumption of unpasteurized apple cider or juice have been reported (4, 5, 6, 8, 13). That none of these investigations was able to definitively identify the mode of contamination underscores the difficulty of doing so. In our investigation, we likewise were unable to definitively know how the apple cider became contaminated. However, three explanations are plausible. First, droppers might have been used earlier, although the orchard owner might have been unaware of the fact. Because the infectious dose of E. coli O157:H7 is relatively low, even a small number of contaminated droppers could cause human illness. At the time of the investigation, the cider producer indicated that he had not yet used droppers to make cider, because the apples had just begun to fall from the trees. This suggests that the practice of using fallen apples was acceptable. Second, cross-contamination might have occurred from an infected asymptomatic worker, and because of the wide clinical spectrum of E. coli O157:H7 infections, an infected worker might have experienced a relatively mild diarrheal illness and continued to work in Orchard A. Third, a worker might have cross-contaminated apples by wearing gloves or other clothing that had been used elsewhere and become contaminated with ruminant feces.
Because the cider operation used potable water obtained from a municipal water system in a rural water district, water contamination was unlikely. Furthermore, no cases of E. coli O157:H7 infection had been reported in the geographic area of the orchard or rural water district during that calendar year.

Although all laboratory tests failed to detect the presence of E. coli O157:H7 in sampled lots of the cider and environmental specimens, this is not unusual, compared with results of other investigations (6, 9, 10, 26). Investigators of a substantial outbreak of E. coli O157:H7 infection and cryptosporidiosis associated with drinking unpasteurized commercial apple juice—British Columbia, California, Colorado, and Washington.-MMWR 45(44):975.

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Note: The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the Centers for Disease Control and Prevention.

REFERENCES


