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Outbreak of Cryptosporidiosis Associated with a Swimming Pool, Auglaize County, Ohio, 2004

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Abstract

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CHAPTER I: LITERATURE REVIEW

Clinical

Human infection caused by *Cryptosporidium* species was first described in 1976 (1); cryptosporidiosis came to prominence in the 1980's as an emerging infectious disease and opportunistic infection in AIDS patients. Cryptosporidiosis generally manifests in the gastrointestinal tract causing symptoms of profuse watery non-bloody diarrhea that can be associated with fever, abdominal pain, and vomiting (2). Cryptosporidiosis is most often an acute, self-limited illness in immunocompetent persons with symptoms lasting 2–3 weeks and an average stool frequency of six times per day (range 2–20) (2, 3). Other presentations among immunocompetent persons include asymptomatic carriage and persistent diarrhea that might last several weeks (4). There was no effective drug treatment for cryptosporidiosis until 2004 when nitazoxanide was approved for immunocompetent patients aged 1–11 years and all patient ≥ 1 year in 2005(5).

Among immunocompromised persons, cryptosporidiosis is most well-defined in AIDS patients. The level of immunosuppression plays a role in how *Cryptosporidium* infection manifests with mildly immunocompromised persons presenting similarly to the immunocompetent (6). However, as immune function declines (particularly CD4 count <200 cells/ml), the resulting *Cryptosporidium* infection begins manifesting as a relapsing, chronic or even cholera-like diarrhea (3). Additionally, immunocompromised persons, particularly those with AIDS, might develop extraintestinal cryptosporidiosis, most commonly in the biliary system (7). Other sites of *Cryptosporidium* infection include the lungs, pancreas, and stomach (6).

Epidemiology

The Centers for Disease Control and Prevention (CDC) collects data on cases of cryptosporidiosis which has been a nationally notifiable condition since 1995. The most recent summary of U.S. cryptosporidiosis cases summarizes data for the years 2009 and 2010. In 2009, 7,656 cases of confirmed and probable cryptosporidiosis cases were reported for a rate of 2.5 per 100,000 population and in 2010, 8,951 confirmed and probable cases were reported for a rate of 2.9 per 100,000 population (8). These numbers were less than the peak year in 2007 when 11,657 cases were reported; however, they do represent a sustained increase in cryptosporidiosis cases since 2004. The most frequently affected age group was children aged 1–9 years followed by adults aged 25–29 years likely reflecting the secondary transmission that occurs from children to their caregivers (8). Surveillance for cryptosporidiosis in the U.S. has shown a consistent seasonality with peak onset occurring in the early summer through early fall months corresponding with the peak time of recreational water activities in the U.S. (8)

Microbiology

There are many species within the genus *Cryptosporidium*, of which *C. hominis* and *C. parvum* cause >95% of human infections.(9) Testing for *Cryptosporidium* is not usually included in routine stool testing for ova and parasites; therefore, clinicians must request *Cryptosporidium* testing specifically which is usually done with acid fast staining of the stool, direct fluorescent antibody (DFA), or enzyme immunoassays for detection of *Cryptosporidium* spp. antigens. These methods of testing identify the genus *Cryptosporidium* only. In order to determine the specific species or genotype of *Cryptosporidium* causing infection, molecular typing must be done. Molecular typing is generally done using small subunit rRNA-based tools. The increasing use of molecular typing of *Cryptosporidium* spp. in outbreak situations has led to improved understanding of outbreak sources (10). Molecular typing of *Cryptosporidium* spp. has also led to improved understanding of its epidemiology. *C. hominis* (previously known as *C. parvum* genotype I) primarily exists in a human-to-human transmission cycle. *C parvum* (previously known as *C. parvum* genotype II) can infect both humans and ruminants (e.g. preweaned calves), each with their own transmission cycle that intersect in zoonotic disease (10).

Cryptosporidium is transmitted by the fecal-oral route in the form of its oocysts. *Cryptosporidium* spp. possess characteristics that make it a public health challenge. First, it can take as few as 9–10 oocysts to cause gastrointestinal infection in healthy persons (11, 12). Additionally, ill persons can excrete up to 10^7–10^8 oocysts in each stool (13). Oocyst excretion has been shown to last up to 2 months after symptoms have resolved with a mean excretion period of 7 days after symptoms have resolved (14). Finally, oocysts are extremely chlorine-tolerant, making this traditional barrier for pathogen transmission in water ineffective for *Cryptosporidium* (15, 16).

Transmission

Contributing to *Cryptosporidium's* challenges to public health are the many mechanisms of transmission: person-to-person, zoonotic foodborne, and waterborne (including both drinking water and recreational water). The person-to-person route is responsible for secondary transmission that occurs during outbreaks when one person is exposed to the outbreak source followed by infection of their contacts(17). Case-control studies have identified contact with children with diarrhea and contact with any person with diarrhea to be significant risk factors for cryptosporidiosis (2, 18). These same case-control studies also identified contact with cattle as a risk factor for cryptosporidiosis. Outbreaks of cryptosporidiosis among those providing veterinary care and children who came into contact with animals in farm and petting-zoo settings demonstrate this form of transmission as well (19-21). Foodborne transmission of *Cryptosporidium* has been documented, with identified outbreak sources including raw produce and food handlers who are ill (22-24). Mechanisms for contamination of food items include treating crops with fertilizer derived from animal or human feces containing oocysts, irrigating with contaminated water, handling of food by food workers with contaminated hands, and packing or storing food on contaminated surfaces (25).

Transmission of *Cryptosporidium* through drinking water came to prominence following the massive 1993 outbreak of cryptosporidiosis in Milwaukee, Wisconsin. This outbreak was estimated to have affected more than 400,000 people and was caused by residential drinking water contaminated with *Cryptosporidium* oocysts (26). It was postulated that the drinking water source water from Lake Michigan may have become contaminated by sewage overflow since the outbreak was caused by the human-human transmitted species *C. hominis* (25). Multiple other outbreaks have occurred in addition to the Milwaukee outbreak with the causes most often a combination of contaminated source water, high turbidity, and failures at the water treatment plant (25). However, changes to surface water treatment regulations following the Milwaukee outbreak requiring methods to inactivate or remove oocysts have caused surface drinking water-related outbreaks to virtually disappear in the U.S. (27).

Transmission through recreational water has increasingly been recognized as the cause of many cryptosporidiosis outbreaks. CDC has collected data regarding recreational water–associated disease outbreaks since 1978. For the reporting period of 1986–1988, a total of 10 gastrointestinal illness outbreaks associated with recreational water were reported, of which none were caused by *Cryptosporidium* (28). The first detected U.S. recreational water–associated cryptosporidiosis outbreak occurred in Los Angeles, CA in 1988 (29). During the 1993–1994 reporting period, 14 gastrointestinal illness outbreaks associated with recreational water were detected, and *Cryptosporidium* became the most common etiologic agent reported, causing 43% of the outbreaks. During this time period, untreated lakes were the most common water exposure (30). During the 1997–1998 reporting period, 9 recreational water–associated outbreaks of cryptosporidiosis were detected, 8 of which were associated with treated venues (31). The report on the surveillance period of 2001–2002 noted a statistically significant increase in recreational water–associated gastrointestinal illness outbreaks since 1993 (14 outbreaks reported in 1993–1994 vs. 30 outbreaks reported in 2001–2002). From 1993–2002, *Cryptosporidium* accounted for more than 65% of outbreaks occurring in treated water venues (32). The increase in recreational water-associated outbreaks was sustained in 2003–2004 and has again continued to increase up through the most recent report of 2007–2008 (33-35).

Several factors have contributed to the rise of cryptosporidiosis outbreaks in recreational water and the ease with which *Cryptosporidium* is transmitted by this vehicle. As described previously, the low infectious dose, high titer of oocysts in stool, and prolonged excretion of oocysts in stool even after resolution of diarrhea are contributing factors to *Cryptosporidium* transmission in recreational water. However, there are additional environmental and human factors that specifically enable *Cryptosporidium* transmission in recreational water. Microbiological assessments of recreational surface water and wastewater have demonstrated that *Cryptosporidium* is a common contaminant (36-38). The ability for oocysts to persist in the environment for months and remain infectious in both fresh water and salt water is one such factor that contributes to the ubiquity of *Cryptosporidium* in the environment (39). Animals also play a role in the contamination of untreated recreational water (e.g., lakes and rivers). During heavy rainfall, animal feces containing oocysts can be washed into untreated recreational water. Once treated recreational water (e.g., pools) becomes contaminated with oocysts, it is difficult to inactivate them. Most treated recreational water venues rely on chlorine as a barrier to the transmission of infectious pathogens, and *Cryptosporidium* oocysts are extremely chlorine-tolerant (15, 16). At currently recommended chlorine levels (1 ppm free chlorine), *Cryptosporidium* oocysts can survive for approximately 10.6 days. For comparison, *E. coli* O157:H7 survives for less than a minute at this chlorine concentration(40). *Cryptosporidium* oocysts' size (approximately 5 micrometers) also allows them to bypass filters typically used for treated recreational water venues. Additional factors also play a major role in the transmission of *Cryptosporidium* through recreational water. First, diarrheal illness in the general population is common. Surveys show that 5% of the general American population has had diarrhea in the preceding month with prevalence as high as 10.3% in children under 5 years (41). A review of 33 studies of acute gastrointestinal illness in developed countries produced an estimate 0.1–3.5 cases of diarrhea per person per year with higher prevalence in children (42). Of these cases of diarrhea, cryptosporidiosis is a common cause accounting for approximately

748,000 cases of diarrhea in the U.S. annually (43). A study of serologic responses to two *Cryptosporidium* antigens among National Health and Nutrition Examination Survey (NHANES) participants found that 13–30% of participants had a serologic response, depending on the type of drinking water they consumed (44). In addition to cryptosporidiosis being common in the general population, swimming is the second most popular sports activity in the United States resulting in more than 300 million swimming visits each year (US Bureau of the Census. 2012 Statistical Abstract of the United States. Recreation and Leisure Activities: Participation in Selected Sports Activities 2009). Given the frequency of cryptosporidiosis in the population and the popularity of swimming, there are multiple opportunities for recreational water to become contaminated with *Cryptosporidium*. Fecal contamination of recreational water is a common event. A community-based survey found that more than 2% of those interviewed were incontinent of feces and that 70% with fecal incontinence were less than 65 years of age (45). A national survey of NHANES participants found an estimated prevalence of fecal incontinence among noninstitutionalized U.S. adults to be 8.3% (46) Concerning fecal incidents in pools, a study which aimed to determine the prevalence of parasites in fecal material from chlorinated swimming pools collected 293 formed stools from 47 pools during one summer swimming season for an average of 6.2 fecal incidents per swimming pool in one season (47). Finally, misconceptions regarding swimming and recreational water as well as common behaviors in the pool put many at risk for acquiring a recreational water–associated illness. Lack of awareness of recreational water–associated illness is a major issue. Focus groups convened by CDC in 1998 and 1999 found that parents did not consider swimming to be the same thing as communal bathing, waterborne disease is something only found in the developing world, and chlorinated water is essentially "sterile" and therefore, cannot transmit disease (48). Additionally, a poll conducted by the National Consumer League in 2004 confirmed the CDC focus group findings as well as documenting that 18% of respondents believed it was acceptable to swim while having diarrhea (National Consumer League. Healthy Pools Survey. Washington, DC: National Consumers League; 2004). These beliefs likely explain why swimmers engage in such behaviors as ingesting pool water. A study of swimmers who actively swam for at least 45 minutes found that adults swallowed, on average, 16 mL (0.5 fluid ounces) of water and children swallowed 37 mL (1.3 fluid ounces) (49). A single diarrheal contamination incident can introduce enough oocysts to a typical treated recreational water venue such that a single mouthful could lead to infection.

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CHAPTER II: MANUSCRIPT

Outbreak of Cryptosporidiosis Associated with a Swimming Pool, Auglaize County, Ohio, 2004

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ABSTRACT

The incidence of recreational water–associated outbreaks in the United States has significantly increased since national surveillance began in 1978. This increase has been driven by outbreaks caused by *Cryptosporidium* and associated with treated recreational water venues (e.g., pools). On August 9, 2004, Auglaize County Health Department staff detected several laboratory-confirmed cases of diarrhea caused by *Cryptosporidium* in a three-county area. Epidemiologic, laboratory, and environmental health methods were used to investigate the outbreak. While the outbreak was investigated and prior to final results being available, proactive prevention measures such as closure and hyperchlorination of pools and a community-wide education campaign were undertaken. The matched case-control study found that cryptosporidiosis was significantly associated with swimming in Pool A (matched odds ratio [mOR] 121.7, 95% confidence interval [CI] 27.4–∞) as were swimming, wading, or entering any swimming pool and swimming in or entering recreational water. Twenty-eight of 45 (62.2%) stool specimens tested were positive for *Cryptosporidium* direct immunofluorescent assay and molecular typing of 18 isolates identified *Cryptosporidium hominis*, subtype IdA15G1. Samples taken from the

deep and shallow ends of Pool A were positive for *Cryptosporidium* spp. by real-time polymerase chain reaction. This report records the first detection of *Cryptosporidium hominis*, subtype IdA15G1 in the United States. The results of the combined methods of this investigation indicate that Pool A was the cause of the outbreak. The findings also suggest a proactive public health response when increased *Cryptosporidium* transmission is detected and before an outbreak source is epidemiologically implicated might prevent a focal cryptosporidiosis outbreak from evolving into a community-wide outbreak.

INTRODUCTION

The incidence of recreational water–associated outbreaks in the United States has significantly increased (negative binomial regression, $p<0.001$) since national surveillance began in 1978 (1). This increase has been driven by outbreaks associated with treated recreational water venues (e.g., pools) and caused by *Cryptosporidium*. The first detected treated recreational water–associated outbreaks of cryptosporidiosis were epidemiologically linked to a learner pool in Doncaster, United Kingdom and a school pool in Los Angeles, United States in 1988 (2-4). During 2007–2008, *Cryptosporidium* caused 59 (72%) of 82 treated recreational water–associated outbreaks with an identified infectious etiology (1). In contrast to chlorine-susceptible pathogens, which are inactivated within minutes, *Cryptosporidium* is extremely chlorine-tolerant with inactivation taking 3.5–10.6 days at typically required or recommended free chlorine levels (i.e., 1–3 mg/L, pH 7.5, temperature 77°F [25°C])(5). Thus, *Cryptosporidium* transmission can occur even in well-maintained treated recreational water venues and a focal outbreak associated with one venue can expand into an outbreak associated with multiple recreational water venues or other community settings (e.g., child care centers) if not adequately contained(6, 7).

The increasing use of molecular typing of *Cryptosporidium* spp. has helped elucidate the epidemiology of cryptosporidiosis (e.g., differentiating clusters during outbreak investigations) (8, 9). There are many species within the genus *Cryptosporidium*, of which *C. hominis* and *C. parvum* cause >95% of human infections (10). *C. hominis* and *C. parvum* are morphologically indistinguishable, thus molecular typing must be done to speciate these isolates. Identifying the *Cryptosporidium* species can provide key epidemiologic clues; for example, *C. hominis* primarily exists in a human-to-human transmission cycle. *C. parvum* can infect both humans and ruminants (e.g., pre-weaned calves), each with their own transmission cycle that intersects in zoonotic disease (11).

On August 9, 2004, Ohio's Auglaize County Health Department (ACHD) detected a laboratory-confirmed case of diarrhea caused by *Cryptosporidium*. After also noting multiple additional cases of cryptosporidiosis in neighboring counties in the Ohio Disease Reporting System, ACHD contacted the health departments of Mercer and Shelby Counties to ask if they had found any common exposures among their reported cases. Preliminary interviews revealed that several additional unreported cases of diarrhea had occurred among Pool A's swim team members. Because this outbreak was detected a few days before a large festival, with >9,000 anticipated attendees, that could potentially lead to expanded transmission, an investigation with , the U.S. Centers for Disease Control and Prevention (CDC) was initiated. The investigation was aimed at determining the magnitude of the outbreak and identifying risk factors for cryptosporidiosis in Auglaize, Shelby, and Mercer Counties.

Previous investigations of cryptosporidiosis outbreaks have demonstrated that interventions to prevent transmission should be initiated as soon as possible, even before the outbreak source is definitively identified (7). Therefore, to limit transmission, proactive prevention measures were concurrently undertaken in the tri-county area. These included creating a press release for the local media and contacting child care facilities, public schools, physician offices, medical facilities, and nursing homes to convey prevention messages. Signage was also created for distribution to local pools reminding pool patrons to not swim while ill with diarrhea and instructing them to use proper handwashing techniques. Additional measures focused on the festival and included teaching proper handwashing techniques to festival food handlers, instructing food handlers to refrain from food preparation if ill, placing signage encouraging handwashing in and around festival restrooms, and strategically locating handwashing stations. Informational packets on cryptosporidiosis and the importance of good hygiene practices when ill were distributed to festival attendees. Suspect Pool A was hyperchlorinated on August 12 and 13 (12). Two neighboring pools were also closed and hyperchlorinated on the same days as Pool A.

METHODS

Case-control study

For this outbreak investigation, a confirmed case was defined as a resident of Auglaize, Mercer, or Shelby Counties, Ohio, who 1) had diarrhea (i.e., ≥3 loose stools in a 24-hour period) in July–August 2004, 2) did not travel to South Bass Island, Ohio, and 3) had laboratory-based evidence of *Cryptosporidium* infection (13). A probable case was defined as a person who met the above criteria but did not have laboratory-based evidence of infection and was epidemiologically linked to confirmed case. The tri-county health departments contacted healthcare providers (e.g., physicians and hospitals) in their respective jurisdictions to encourage testing of symptomatic persons and reporting of positive test results. Based on reports called into the tri-county health departments, a line list of case-patients was generated.

To identify community risk factors for cryptosporidiosis, a case-control study was conducted. Case-patients thought to be the first ill in each household were recruited from the aforementioned line list. Two controls, who reported no gastrointestinal illness in July–August 2004, were matched to each case-patient by age range (i.e., 0–<2 years, 2–<6 years, 6–<18 years, and ≥18 years) and county of residence. Area phone books were used to identify potential controls. A person listed on the first page of each section was selected at random and then that person and subsequently every tenth listed person was recruited for the study. As with the case-patients, only one person per household was interviewed as a control. For study participants under the age of 18 years, a parent or guardian answered questions on behalf of the child or the parent's or guardian's permission was obtained prior to speaking with the child (child 12 years or older). Prior to the start of the interview, the interviewer attempted to verify that the case was the first ill in the household.

Case-patients and controls were administered a standardized questionnaire asking about exposures in the 2 weeks prior to the onset of diarrhea in the case-patient. Questions about the following were included: food and drinking water consumption; recreational water, child care, household, farm, and animal exposures; attendance of events with ≥50 persons; and travel history. Because hypothesis-generating interviews indicated a common recreational water exposure among case-patients, detailed questions were asked about recreational water activities in general and at specific pools, including Pool A. Additionally, data were collected on the symptoms case-patients experienced and time lost from work and from activities of daily living due to illness. All data analyses were performed using SAS, version 9.3 (SAS Institute Inc., USA). Odds ratios (OR) were calculated using conditional logistic regression to account for matching. Ninety-five percent confidence intervals that did not contain the null value were considered significant.

Laboratory investigation

Stool testing for *Cryptosporidium* was requested of those individuals who were experiencing diarrhea or whose diarrhea had resolved within the last two weeks. Testing for *Cryptosporidium* was performed at the Ohio Department of Health (ODH) laboratory by direct immunofluorescent assay (DFA) and results were confirmed by follow-up DFA at CDC. *Cryptosporidium* isolates were subtyped at CDC using DNA sequence analysis (14). Stool specimens from a subset of initially identified case-patients were tested by ODH for bacterial and viral pathogens to rule out bacterial and viral etiologies of diarrheal illness.

Environmental health investigation

The environmental health investigation focused on Pool A, which was anecdotally implicated early in the outbreak investigation. Maintenance records and daily attendance numbers were reviewed for the months of July and August. On August 11, 2004, the pool was closed and a private company took water samples from the wading pool and the deep and shallow ends of the main pool. These samples were processed by the ODH laboratory using U.S. Environmental Protection Agency Method 1622 (15). The resulting immunomagnetic separation beads were sent to CDC for molecular analysis (16). In addition to the water sampling, aquatic staff were interviewed, using a standardized questionnaire asking about whether or not they had diarrhea, their activities and duties at the pool, and observed pool patron behavior in July–August 2004.

RESULTS

Case-control study

Sixty-three case-patients were recruited for the case-control study. Of these, four (5.9%) refused to participate in the study; two (2.9%) case-patients traveled to South Bass Island, Ohio and were potentially associated with another outbreak of gastrointestinal illness; and one (1.5%) case-patient reported travel to an unknown destination (i.e., might have traveled to South Bass Island).

Of the 56 enrolled case-patients (median age=8.0 years; range: 1.3–61 years), 49 (87.5%) were children and 27 (48.2%) were male. The most commonly reported symptoms were diarrhea, tiredness/fatigue, loss of appetite, and abdominal cramps (Table 1). Fifty-one case-patients reportedly experienced a median maximum of 7 stools (range: 3–25 stools) in 24 hours and diarrhea lasting a median of 6 days (range: 1–19 days). Of 53 patients, 33 (62.3%) reportedly experienced waxing and waning of diarrhea that occurred a median of 2 times (range: 1–8 times) before complete resolution. No case-patients reported being immunocompromised. Three case-patients were reported to be seen in an emergency department; two were reported to be hospitalized. Of 56 case-patients or case-patients' parent or guardian, 36 (64.3%) (4 [11.1%] adult case-patients, 32 [88.9%] case-patients' parent or guardian) reported being employed at the time of illness; 12 (33.3%) (2 [16.7%] case-patients, 10 [83.3%] case-patients' parent or guardian) of them reported missing work because of the illness for a median of 2 days (range: 1–14 days). Almost two thirds (36 [64.3%] of 56) of case-patients (6 [16.7%]) or case-patients' parent or guardian (30 [83.3%]) were reportedly not able to perform activities of daily living because of their illness or their child's illness for a median of 4 days (range: 1–17 days). Three (5.4%) case-patients reportedly continued to do recreational water activities while they had diarrhea and 10 (17.9%) case-patients reportedly participated in recreational water activities within two weeks of their diarrhea completely resolving. The peak onset of symptoms occurred on August 8 (Figure).

One case-patient included in the above descriptive analysis lacked properly matched controls and so was excluded from matched case-control analysis. The number of males and females did not differ significantly between cases and controls. Findings of bivariate analyses of data on individual exposures are summarized in Table 2. Cryptosporidiosis was significantly associated with swimming in Pool A (matched odds ratio [mOR] 121.7, 95% confidence interval [CI] 27.4–∞) as were swimming in any swimming pool or recreational water. Having an adult household contact (mOR 5.4, 95% CI 1.6–23.0) and a child household contact ill with diarrhea in the 2 weeks prior to symptom onset (mOR 41.2, 95% CI 6.7–>999) were also significant risk factors for cryptosporidiosis. When case-patients who reported any ill household contacts in the 2 weeks prior to symptom onset were excluded from the analysis, swimming in Pool A remained a significant risk factor (mOR 49.6, 95% CI 3.0–∞) as did swimming in any swimming pool (mOR 19.6, 95% CI 4.1–∞) or recreational water (mOR 14.6, 95% CI 3.0–∞).

Consumption of cider or juice; contact with a cat or dog, farm animal, or animal waste or manure; and swimming in a private/residential pool were significantly protective. More than 95.0% (22/23) of controls who swam in a private/residential pool did not swim in Pool A. When these exposures were individually modeled as predictors along with swimming in Pool A, they remained protective but were no longer statistically significant.

Laboratory investigation

Forty-five stool specimens were tested by CDC; 28 (62.2%) were positive for *Cryptosporidium* by DFA. Molecular typing of 18 isolates identified *Cryptosporidium hominis*, subtype IdA15G1. Stool specimens from 25 initial case-patients were tested for the presence of bacterial and viral pathogens, of which 24 (96.0%) were negative; one (4.0%) had no growth, including normal intestinal flora, and thus was considered an invalid clinical specimen.

Environmental health investigation

Pool A consisted of a main pool and a wading pool. The main pool had a spread finger layout and areas with a diving well, water slide, kiddie slide, rain drop water feature, and zero-depth entry. The main pool had a capacity of 262,713 gallons. Disinfection of the main pool was achieved through treatment with sodium hypochlorite (NaOCl) delivered by automatic controllers and two sand filters with a required flow rate of 547 gallons per minute. The wading pool had a capacity of 3,093 gallons and a depth of 1–1.5 feet. It was on a separate circulation system and was also disinfected with NaOCl and a sand filter.

The samples taken from the deep and shallow ends of the main pool were positive for *Cryptosporidium* spp. by real-time PCR, while the wading pool samples were negative. Conventional PCR assays for the water samples were negative, so molecular typing of the pool isolates was not possible

A review of Pool A attendance records showed a peak in pool attendance on August 2 (Figure). Further breakdown of the types of patrons in attendance revealed a peak in attendance by pre-school–aged children on August 2. During the pool inspection conducted on August 18, Auglaize County sanitarians noted no violations. Of 38 pool employees, 32 (84.2%) were interviewed. All interviewed pool employees were lifeguards. Eighteen (56.3%) were also swim instructors; 3 (9.4%) also served as managers. The median age of the employees was 17 years (range: 15–21 years). Eight (25.8%) of 31 employees reported having diarrhea since July 1, 2004; two (25.0%) of the eight reported that they continued to go into the water in any pool or participated in other recreational water-related activities while ill with diarrhea. Four (50.0%) of the eight employees reported that they went into Pool A or participated in other recreational water-related activities within the two weeks after their diarrhea completely resolved. Eight (25.0%) of 32 reported observing mothers changing diapers in the area around the wading pool; four (13.3%) of 30 reported observing mothers changing diapers around the main pool. No employees reported observing mothers cleaning or rinsing their child's bottom in the wading or main pool. Fifteen (48.4%) of 31 employees reported fecal incidents in the main pool and 3 (10.0%) of 30 reported fecal incidents in the wading pool during July and August. When the managers were asked what percentage of babies swam in the wading pool versus the main pool, they reported that the majority of babies were in the main pool on a typical day.

DISCUSSION

Through a collaborative epidemiologic, environmental health, and laboratory investigation, Pool A was identified as the source of this outbreak. The epidemic curve for the case-control study displays a single peak in illness onset and suggests a point-source exposure. The case-control study confirmed an association between cryptosporidiosis and swimming, specifically in Pool A. The environmental health investigation through inspection and review of pool records revealed that Pool A was properly operated based on state pool code and peak attendance by pre-school–aged children occurred one incubation period (i.e., 6 days) before the peak in illness onset among study case-patients. Of note, water samples taken from the main pool were positive for *Cryptosporidium* and multiple clinical specimens were positive for *Cryptosporidium hominis*, subtype IdA15G1 indicating a human to human transmission cycle. This subtype was first identified in the stool specimens of human patients in Australia; results were later confirmed locally (17, 18). It appears to be a common anthroponotic subtype in India, Pakistan, and Bangladesh (19-22). This report records the first detection of IdA15G1 in the United States.

Contamination of Pool A on the day of peak attendance by pre-school–aged children could have potentially provided an opportunity for the outbreak to expand in the community with infected persons transmitting *Cryptosporidium* in other public settings (e.g., other recreational water venues and child care facilities). However, analysis of study data did not find a significant positive association between illness and swimming at any other recreational water venue or child care facilities. Pool A and two nearby pools were rapidly closed to swimmers, hyperchlorinated (i.e., remediated to achieve 3-log inactivation of *Cryptosporidium*), and reopened within a few days of the first outbreak-related cases being reported (Figure). This proactive response (i.e., before the case-control study was conducted, the data were analyzed, and the outbreak source was statistically identified) by the pool management and tri-county health departments combined with community education about cryptosporidiosis prevention could be responsible for prevention of community-wide spread of *Cryptosporidium*. Almost 95% of case-patients reported not swimming while ill with diarrhea. This healthy swimming behavior is key given that swimmers introduce *Cryptosporidium* to treated recreational water venues and once contaminated, even a properly maintained pool, such as Pool A, can transmit *Cryptosporidium*. In contrast, 25% of pool employees reported entering the water while ill with diarrhea. Ill pool employees might have found it difficult to abstain from entering the water while performing work duties without admitting to being ill. Pool managers should institute policies that allow employees to call out sick or perform alternate duties that do not require entering the water while they are ill with diarrhea. Such policies would be analogous to those in the food service industry that deter employees from handling food while ill with diarrhea.

A previously reported study suggests that a healthy swimming education campaign, such as the one conducted in the days following the detection of this outbreak, may help prevent cryptosporidiosis associated with treated recreational water (23). Following a massive statewide outbreak of cryptosporidiosis in Utah associated with recreational water in 2007, local and state public health officials disseminated healthy swimming messages via the web, television advertisements, public service radio announcements, and posters at pools before the 2008 summer swim season (6). Healthy swimming messages were also targeted at schools, competitive water sports teams, and child care facilities. A 2009 national survey found that Utah residents were significantly more likely to know not to swim while ill with diarrhea than residents of other states (100% vs. 78.4%). No recreational water–associated outbreaks were detected in Utah during 2008–2011.

Analyses of study data also found that having an adult or child household contact ill with diarrhea in the 2 weeks prior to illness onset was also associated with disease. While this finding would not be expected given that the investigators attempted to interview the first ill in the household, when asked about others ill in the household, it was not clear whether respondents were answering that others were ill at the same time or whether they were ill before the case-patient (i.e., the case-patient might not have actually been the first ill in the household). Having ill household contacts has been identified as a risk factor for cryptosporidiosis in multiple studies and is a major mechanism for secondary transmission (6, 24-26). While the rapid response to this outbreak provides a model for prevention of community-wide transmission of *Cryptosporidium*, transmission still occurred within households and highlights the need for education regarding basic personal hygiene such as handwashing after toileting, assisting others with toileting, or changing a diaper and before preparing food. Handwashing is a simple intervention that can not only effectively prevent transmission of *Cryptosporidium*, but also other infectious etiologies of gastrointestinal and respiratory illness (27-29).

There were also several exposures evaluated in the case-control study that were found to be significantly protective in bivariate analysis. The protective effect of contact with animals and animal waste could be a marker for previous *Cryptosporidium* infection, which results in persistently elevated antibody levels that protect against clinical illness (i.e., re-infected individuals are less likely to meet a clinical case definition) (30). Consumption of cider (i.e., made with fecally-contaminated apples picked off the ground vs. the tree) has been previously associated with cryptosporidiosis outbreaks and might also be a marker of previous *Cryptosporidium* infection (8). The protective effect found from swimming in a private residential pool was likely seen because swimming in a private residential pool provided an alternative to swimming in Pool A. Additionally, private residential pools have fewer swimmers and, therefore, may be less likely to become contaminated. Another case-control study of sporadic cryptosporidiosis found residential pool swimming to be protective in univariate analysis although it was not significant in the final multivariate model (25). This hypothesis is supported by the fact that >95.0% controls who swam in a private residential pool did not swim in Pool A.

This investigation had multiple limitations. First, while the investigation aimed to interview the first person in the household who became ill with diarrhea, confusion on the part of both interviewers and respondents regarding the household contact question asking about onset of diarrheal illness in household members potentially resulted in 31/55 interviews of case-patients who were not the first to become ill in the household. This limitation led to the identification of household risk factors for disease instead of those in the community. However, when the 31 secondary case-patients were removed from the analysis, further limiting statistical power, Pool A remained significantly associated with illness. Alternatively, while the interviewed case-patient might not have been the first to become ill in the household, he or she might have had the same exposure as the household member who became ill first (i.e., the interviewed case-patient became ill within hours or a day or two of the first ill). Second there was possible recall bias among study participants given that the questionnaire was administered a few weeks after the outbreak started. Case-patients might have been able to more accurately report their exposures during the two weeks before their illness than controls who did not have an illness to enhance their memory of the time period of interest. Finally, the methods of this outbreak investigation cannot prove that the proactive disease prevention measures taken caused this to remain a focal outbreak. However, the causation criteria of temporality, plausibility, and specificity are met and support the conclusion that the proactive measures taken in this outbreak could have resulted in its containment.

The findings of this investigation suggest that a proactive public health response when increased *Cryptosporidium* transmission is detected and before an outbreak source is epidemiologically implicated (i.e., interventions such as prompt pool closure, hyperchlorination, and reopening) and community education about cryptosporidiosis prevention (i.e., don’t swim when ill with diarrhea) might prevent a focal cryptosporidiosis outbreak from expanding into a community-wide outbreak. However, it also demonstrates that by the time increased transmission is detected it is likely to be too late to prevent household transmission. Historically, cryptosporidiosis outbreak investigations and the ensuing case-control studies focus on community exposures rather than household exposures and secondary transmission. The proactive disease prevention measures taken in this outbreak can do little to prevent secondary transmission within households. Prevention of household transmission requires continual hygiene education before outbreak exposure occurs. This outbreak also highlights the need to work with recreational water venues to develop policies that allow venue staff to perform alternative work duties (i.e., working the front desk) while they are ill with diarrhea. Given the prolonged nature of diarrhea, time lost from work and activities of daily living, and healthcare costs associated with emergency department treatment and hospitalization, disseminating cryptosporidiosis prevention messages (e.g., particularly low-cost measures like good handwashing practices) could be a worthwhile investment (31).

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Table 1. Clinical symptoms reported for case-patients recruited for case-control study

	Case-patients (*N*=56)	
Symptom	*n**	%
Diarrhea	56/56	100
Tiredness/Fatigue	52/55	94.6
Loss of appetite	48/54	88.9
Abdominal cramps	49/56	87.5
Nausea	38/53	71.7
Headache	35/51	68.6
Vomiting	36/55	65.5
Body/Muscle aches	30/50	60.0
Bloating/Gas	29/50	58.0
Fever	32/55	58.2
Weight loss	24/47	51.1
Blood in stool	0/54	0

*Denominator varies due to missing, unknown, or refused values.

Table 2. Bivariate conditional analysis of exposures in case-patients and controls

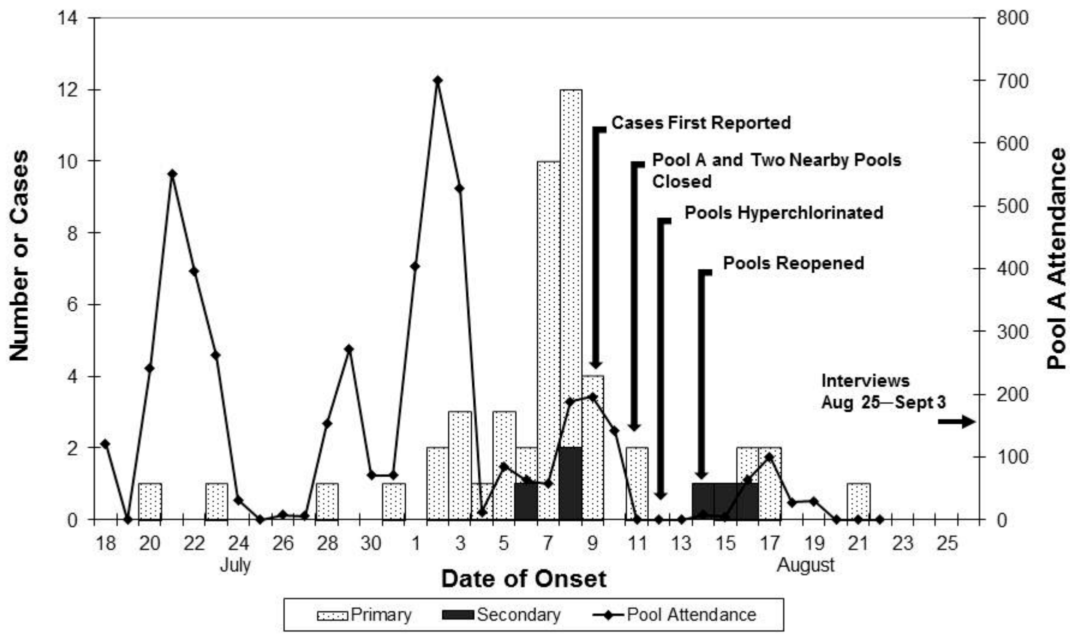
	Case-patients (N=55)		Controls (N=105)		mOR (95% CI)
Exposure	n*	%	n*	%	
Demographics					
Gender (%male)	26/55	47.3	45/98	45.9	1.09 (0.52–2.31)
Human contact					
Adult household contact ill with diarrhea in 2 weeks before symptom onset	13/55	23.6	4/82	4.9	**5.37 (1.61–22.98)**
Child household contact ill with diarrhea in 2 weeks before symptom onset	26/55	47.3	3/94	3.2	**41.18 (6.68–>999)**
Any household contact ill with diarrhea	31/55	56.4	6/83	7.2	10.50 (3.58–41.85)
Child household contact in diapers	14/47	29.8	6/52	11.5	2.59 (0.79–9.88)
Provide childcare in any setting	11/54	20.4	18/84	21.4	1.00 (0.37–2.54)
Contact with any child in diapers	18/50	36.0	43/101	42.6	0.69 (0.30–1.53)
Change any diapers	4/49	8.2	7/100	7.0	1.27 (0.24–6.13)
Contact with anyone who diarrhea	3/51	5.9	11/91	12.1	0.55 (0.09–2.24)
Provide direct care to person with diarrhea	1/52	1.9	2/92	2.2	0.50 (0.01–39.25)
Dietary exposures					
Lettuce or garden salad	22/51	43.1	57/102	55.9	0.56 (0.25–1.25)
Cold cuts, chicken, egg, or tuna salad	29/55	52.7	65/104	62.5	0.69 (0.33–1.43)
Other cold salads (e.g.,potato salad)	8/53	15.1	30/101	29.7	0.49 (0.18–1.19)
Raw vegetables	29/54	53.7	72/105	68.6	0.58 (0.28–1.20)
Raw berries	19/54	35.2	51/102	50.0	0.58 (0.27–1.20)
Raw fruits with skin or peel	35/55	63.6	77/104	74.0	0.63 (0.29–1.34)
Cider or juice	25/55	45.5	72/104	69.2	**0.34 (0.15–0.74)**
Raw shellfish	0/55	0	2/105	1.9	0.83 (0–6.94)
Cooked shellfish	1/55	1.8	5/105	4.8	0.40 (0.01–3.58)
Unpasteurized milk	0/55	0	2/105	1.9	0.83 (0–6.94)
Unpasteurized apple juice or cider	0/55	0	0/105	0	--
Other unpasteurized juices	1/55	1.8	0/104	0	2.00 (0.11–∞)
Unpasteurized cheese	0/55	0	1/105	0.9	2.00 (0–38.0)
Drinking water exposures					
Does not drink water at home	10/52	19.2	9/105	8.6	2.57 (0.85–8.13)
Municipal or city water direct from tap	22/52	42.3	54/104	51.9	0.67 (0.33–1.35)
Municipal or city water with	13/53	24.5	25/104	24.0	0.97 (0.39–2.36)

additional filtration or treatment					
Refrigerator dispenser	19/52	36.5	28/105	26.7	1.69 (0.72–3.97)
Private well water	21/52	40.4	31/104	29.8	1.90 (0.83–4.48)
Private well water with additional filtration or treatment	12/40	30.0	20/105	19.1	2.23 (0.81–6.51)
Commercially bottled water	36/54	66.7	76/104	73.1	0.69 (0.30–1.58)
Do not use ice	13/51	25.5	21/102	20.6	1.37 (0.53–3.56)
Use ice from home	40/53	75.5	76/102	74.5	1.00 (0.20–1.13)
Use ice from outside home	16/50	32.0	48/100	48.0	0.50 (0.20–1.13)
Drink any untreated water from lake, river, or stream	3/53	5.7	6/103	5.8	0.92 (0.15–4.35)
Recreational water exposures					
Swim or enter recreational water	54/55	98.2	62/104	59.6	**43.30 (9.50–∞)**
Lake, pond, river, or stream	7/52	13.5	22/103	21.4	0.43 (0.12–1.26)
Hot tub, spa, whirlpool, jacuzzi	3/54	5.6	7/103	6.8	0.91 (0.13–4.91)
Recreational water park other than swimming pools	6/55	10.9	8/103	7.8	1.59 (0.38–6.96)
Swim, wade in, or enter a swimming pool	53/55	96.4	53/103	51.5	**48.38 (10.70–∞)**
Pool A	49/55	89.1	6/103	5.8	121.67 (27.42–∞)
Pool B	1/55	1.8	4/103	3.9	0.50 (0.01–5.05)
Pool C	5/55	9.1	4/103	3.9	3.68 (0.56–40.46)
Pool D	0/54	0	3/103	2.9	0.52 (0–3.43)
Pool E	3/55	5.5	1/103	1.0	4.37 (0.34–236.16)
Private/residential pool	3/55	5.5	23/103	22.3	0.22 (0.04–0.78)
Hotel/motel pool	3/55	5.5	9/103	8.7	0.64 (0.11–2.78)
Attend large social event (≥50 people)	41/54	75.9	81/104	77.9	0.86 (0.35–2.17)
Travel					
To state fair	2/54	3.7	7/103	6.8	0.57 (0.06–3.00)
Outside of counties, other than South Bass Island or fair	21/53	39.6	48/102	47.1	0.67 (0.30–1.46)
Animal contact					
Calf	3/49	6.1	12/94	12.8	0.37 (0.04–1.76)
Cat or dog	35/53	66.0	88/103	85.4	0.30 (0.11–0.76)
Farm animal contact	12/50	24.0	43/98	43.9	0.41 (0.16–0.94)
Other animal contact (amphibians, reptiles, other)	4/49	8.2	19/97	19.6	0.41 (0.10–1.36)
Animal waste/manure	11/49	22.5	41/99	41.4	0.35 (0.13–0.85)
Visit, live, or work on a farm	12/54	22.2	27/103	26.2	0.81 (0.35–1.80)

mOR, Matched odds ratio; CI, confidence interval.

*Denominator varies due to missing or unknown responses.

Figure. Number of primary and secondary* cryptosporidiosis cases by date of illness onset and pool attendance — Auglaize County, Ohio, July–August 2004 (N=55)



*Defined as cases who did not swim in Pool A

CHAPTER III: SUMMARY, PUBLIC HEALTH IMPLICATIONS, AND POSSIBLE FUTURE DIRECTIONS

Through a collaborative epidemiologic, environmental health, and laboratory investigation, Pool A was identified as the source of this outbreak. The epidemic curve for the case-control study displays a single peak in illness onset and suggests a point-source exposure. The case-control study confirmed an association between cryptosporidiosis and swimming, specifically in Pool A. The environmental health investigation through inspection and review of pool records revealed that Pool A was properly operated based on state pool code and peak attendance by pre-school–aged children occurred one incubation period (i.e., 6 days) before the peak in illness onset among study case-patients. Of note, water samples taken from the main pool were positive for *Cryptosporidium* and multiple clinical specimens were positive for *Cryptosporidium hominis*, subtype IdA15G1 indicating a human to human transmission cycle. This report records the first detection of IdA15G1 in the United States. Contamination of Pool A on the day of peak attendance by pre-school–aged children provided an opportunity for the outbreak to expand in the community with infected persons transmitting *Cryptosporidium* in other public settings (e.g., other recreational water venues and child care facilities). However, analysis of study data did not find a significant positive association between illness and swimming at any other recreational water venue or child care facilities. Pool A and two nearby pools were closed to swimmers, hyperchlorinated (i.e., remediated to achieve 3-log inactivation of *Cryptosporidium*), and reopened within a few days of the first outbreak-related cases being reported. This proactive response (i.e., before the case-control study was conducted; the data were analyzed; and the outbreak source was statistically identified) by the pool management and tri-county health departments combined with community education about cryptosporidiosis prevention could be responsible for prevention of community-wide transmission of *Cryptosporidium.*

Swimming is a popular recreational activity: in the United States, during 2009, there were over 300 million swimming visits by persons over the age of six years (U.S. Bureau of the Census. 2012 Statistical Abstract of the United States. Recreation and Leisure Activities: Participation in Selected Sports Activities 2009). Swimming is also an excellent form of aerobic physical activity and should be encouraged. In order for swimmers to enjoy this activity in a healthy environment, state and local public health agencies and the aquatics sector should collaborate and take a multi-pronged approach to preventing the transmission of the parasite in treated recreational water venues.

Healthy swimming behaviors are the mainstay of recreational water–associated outbreak prevention, the most important of which is for swimmers to avoid swimming when they have diarrhea. Recreational water venues should institute and enforce diarrhea exclusion policies especially for children who are not yet toilet trained. Recreational water venues should also have a policy to allow staff who are ill with diarrhea to refrain from work duties that involve entering the water. A policy like this might have further optimized prevention of this Ohio outbreak as half of the pool staff participated in water activities while they were ill with diarrhea.

Swimmers can also take measures before entering the pool to reduce the risk for disease transmission including washing with soap, particularly the perianal area, before entering the water. Caregivers should also wash young children with soap and water prior to entering the water. Recreational water venues can facilitate these behaviors by providing an adequate number of clean, functioning, and easily accessible facilities with showers, toilets, and sinks stocked with soap. Caregivers can minimize water contamination by having their children take frequent bathroom breaks and checking diapers every 30–60 minutes. Recreational water venues can also provide diaper-changing facilities with handwashing stations located near the water to facilitate frequent diaper changes. Surveys have found that there is a lack of knowledge about recreational water illness in the swimming public (National Consumer League. Healthy Pools Survey. Washington, DC: National Consumers League; 2004). Beliefs that chlorine creates a sterile environment, waterborne illness is not a problem in the developed world, and swimming while ill with diarrhea is acceptable all point to the public's need for healthy swimming education. To make swimmers aware of recreational water illness and encourage the healthy swimming behaviors that might have prevented the Auglaize County, Ohio outbreak, federal, state and local public health agencies should create healthy swimming education campaigns. The week before the Memorial Day holiday in the United States has been designated Recreational Water Illness and Injury (RWII) Prevention Week (www.cdc.gov/healthywater/swimming/rwi/rwi-prevention-week/index.html). This week, leading up to the kick-off of the summer swimming season in the United States, is an opportunity to promote healthy swimming behaviors. Methods used by CDC to promote RWII Prevention Week include release of a Morbidity and Mortality Weekly Report (MMWR) on an aspect of recreational water illness prevention, and, in 2013, press releases from both CDC and a template that can be used by state and local health departments, features on the main CDC website, social media outreach via Facebook posts and Twitter chats, and creation of a healthy swimming mobile application. RWII

Prevention Week has been observed since 2005 and the activities associated with this observance week have been increasingly adopted by state and local health agencies with the goal of increasing awareness and knowledge about healthy swimming behaviors, and ultimately having the swimming public adopt those behaviors.

Another tool for use by state and local public health agencies as well as the aquatics sector to reduce the burden of recreational water–associated illness is the Model Aquatic Health Code (www.cdc.gov/healthywater/swimming /pools/mahc/). In the U.S., there is no federal regulatory authority responsible for treated aquatic facilities (e.g. swimming pools, water parks, etc.); and requirements for preventing and responding to recreational water-associated illnesses and injuries vary among state and local agencies. The Model Aquatic Health Code (MAHC) is intended to be a data-driven, best practices-based, risk reduction tool that can be used by state and local jurisdictions to prevent disease and injuries and promote healthy and safe recreational water experiences. The MAHC includes modules on the design, construction, operation, and maintenance of public swimming pools and other aquatic facilities. Use of the MAHC is intended to decrease incidence of recreational water-associated illness; decrease injuries and deaths from drowning; and decrease swimming-related emergency department visits for pool chemical-associated health events. Furthermore, adoption of the MAHC will also encourage other improvements in public health such as collection of more meaningful inspection data that can be used to track progress and identify areas warranting further action; enhanced ability to assess and respond to trends on a national and regional scale; and development of a research agenda to address gaps.

Cryptosporidium's extreme chlorine tolerance calls for alternative methods for disinfection of treated recreational water venues. Two such methods include ozone and ultraviolet disinfection systems (1-5). These supplemental/secondary disinfection systems have been developed for use in recreational water venues and are able to inactivate *Cryptosporidium* oocysts. While technology can address some aspects of *Cryptosporidium* transmission, ultimately, it is simple hygiene messages that will keep infectious pathogens out of the water in the first place. Prevention of household transmission requires hygiene education before outbreak exposure occurs. Given the prolonged nature of diarrhea, time lost from work and activities of daily living, and healthcare costs associated with emergency department treatment and hospitalization, disseminating cryptosporidiosis prevention messages (e.g., particularly low-cost measures like good handwashing practices) is a worthwhile investment(6).

1. Betancourt WQ, Rose JB. Drinking water treatment processes for removal of Cryptosporidium and Giardia. *Vet Parasitol* 2004;126(1-2):219-34.

2. Craik SA, Weldon D, Finch GR, et al. Inactivation of Cryptosporidium parvum oocysts using medium- and low-pressure ultraviolet radiation. *Water Res* 2001;35(6):1387-98.

3. Rochelle PA, Upton SJ, Montelone BA, et al. The response of Cryptosporidium parvum to UV light. *Trends Parasitol* 2005;21(2):81-7.

4. Corona-Vasquez B, Samuelson A, Rennecker JL, et al. Inactivation of Cryptosporidium parvum oocysts with ozone and free chlorine. *Water Res* 2002;36(16):4053-63.

5. Korich DG, Mead JR, Madore MS, et al. Effects of ozone, chlorine dioxide, chlorine, and monochloramine on Cryptosporidium parvum oocyst viability. *Appl Environ Microbiol* 1990;56(5):1423-8.

6. Collier SA, Stockman LJ, Hicks LA, et al. Direct healthcare costs of selected diseases primarily or partially transmitted by water. *Epidemiol Infect* 2012;140(11):2003-

APPENDIX

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DATE: June 5, 2012

RE: Determination: No IRB Review Required
Title: Outbreak of Cryptosporidiosis Associated with a Community Swimming Pool, Auglaize County, Ohio, 2004
PI: Jennifer Rittenhouse Cope, MD

Dear Dr. Cope:

Thank you for requesting a determination from our office about the above-referenced project. Based on our review of the materials you provided, we have determined that it does not require IRB review because it does not meet the definition(s) of “research” involving “human subjects” or the definition of “clinical investigation” as set forth in Emory policies and procedures and federal rules, if applicable. Specifically, in this project, you will conduct a secondary data analysis of a de-identified dataset that contains data collected during the Auglaize County Cryptosporidiosis Outbreak associated with a community swimming pool, in 2004. The purpose of this study is to describe the extent of the outbreak, to identify risk factors for Cryptosporidium in Auglaize, Shelby, and Mercer counties, to aid in the development of interventions to control future outbreaks of cryptosporidiosis, and to assist in the development of effective education messages and control measures for the public and for health care providers. Additionally, this outbreak was unique in that it was controlled quickly and it provides an opportunity to examine what was done correctly to control the spread of infection and apply these strategies in future cryptosporidiosis outbreaks. The PI will receive the de-identified dataset from the Waterborne Disease Prevention Branch at the Centers for Disease Control and Prevention. The PI will not have access to any identifiers or coded-links to identifiers now or in the future.

Please note that this determination does not mean that you cannot publish the results. If you have questions about this issue, please contact me.

This determination could be affected by substantive changes in the study design, subject populations, or identifiability of data. If the project changes in any substantive way, please contact our office for clarification.

Thank you for consulting the IRB.

Sincerely,

Carol Corkran, MPH, CIP
Senior Research Protocol Analyst
This letter has been digitally signed