Abstract

Background: An increase in notifications of cryptosporidiosis was observed in Victoria between March and April 2015. Cases mostly resided in one metropolitan region and hypothesis-generating interviews identified common exposures to aquatic facilities. We conducted a case-control study to determine exposure source(s) and facilitate control measures.

Methods: Laboratory-confirmed cases of cryptosporidiosis from the region of interest notified between 1 March and 23 April 2015 were included. Controls residing in the same region were recruited from participants in a population health survey and frequency matched (2 per case) by age group. Details of exposure to potential risk factors were collected using a standardised telephone questionnaire for the 14-days prior to illness for cases, and an analogous exposure period for controls. Univariable and multivariable logistic regression were used to determine risk factors associated with illness using STATA SE 13.1.

Results: Thirty cases and 66 controls were included in the study. Half the cases were less than 12 years of age and 62% were female. Illness was most strongly associated with recreational water exposure at any waterpark (adjusted odds ratio (aOR)=73.5; 95% confidence interval (CI):6.74–802), and specifically at Victorian waterparks (aOR=45.6; 95% CI:5.20–399). Cases were linked with attendance at either a waterpark in the region or an adjacent region. As a result of this investigation, hyperchlorination was completed at identified facilities and swim hygiene information distributed.

Conclusion: This study reinforces the potential for recreational water facilities, particularly waterparks, to act as a transmission source of Cryptosporidium infections. Continued communication to patrons is required to ensure healthy swimming practice in Victorian aquatic facilities.

Keywords: cryptosporidiosis, Cryptosporidium, case-control study, waterborne disease

Introduction

Cryptosporidiosis is a gastrointestinal illness caused by a species of the parasite Cryptosporidium.
Methods

Epidemiological investigation

We identified cases of cryptosporidiosis from the Victorian DHHS communicable disease surveillance system. For the case-control study, cases were defined as a person with laboratory-definitive evidence of Cryptosporidium spp. infection, who resided in the metropolitan region of interest whose onset of diarrhoea (defined as 3 or more loose stools in a 24-hour period) was between 17 February and 21 March 2015. Cases were excluded from the study if they were unable to be contacted after 6 attempts, were unable to recall the date of onset of their diarrhoea at interview or reported international travel for the entire 14 days prior to the onset of their illness. Controls who resided in the same region as cases were recruited from participants in the DHHS annual population health survey. Controls were recruited after all case interviews had been completed. Two controls were recruited per case and were frequency matched by age group to cases (≤5 years, 6–12 years, 13–19 years, 20–29 years, 30–39 years, and 40–49 years). Controls were randomly allocated a 14-day exposure period, analogous to the exposure date range for all cases. Controls were excluded from interview if they had a gastrointestinal illness during their exposure period or reported international travel for the entire exposure period.

A standardised telephone questionnaire was developed and used to collect exposure information on potential risk factors for Cryptosporidium infection in the 14 days prior to the case’s illness, or the assigned 14-day exposure period for controls, with reference to a calendar to aid recall. The risk factor information included person-to-person contact (household, childcare or high-risk occupations), drinking water sources, contact with pets, farm animals and manure and consumption of unpasteurised milk or milk products. Specific recreational water exposures explored were: public or private swimming pools; waterparks; public or private spas; home paddling pools; or any natural bodies of water. In addition, the questionnaire sought clinical information for cases. If the case or control was under 18 years of age, the parent, guardian or another person in the household most familiar with their routine was interviewed.

Univariable and multivariable regression analyses were used to determine risk factors significantly associated with illness after accounting for collinearity in exposure classification, with the strength of an association assessed by estimating age-adjusted Mantel-Haenszel odds ratios, 95% confidence intervals and 2-sided Fisher’s exact probabilities. Statistical tests for associations were determined to be significant when \( P < 0.05 \). All data cleaning and analysis were completed using STATA/SE 13.1 (StataCorp, Texas, USA). In order to focus our analysis to identify unknown primary risk factors for illness, due to the high transmissibility of cryptosporidiosis and the ease of person-to-person spread within a household, we deemed a secondary case to be any case who had a household member who was also a confirmed case in this study but had an earlier onset date of more than 1 incubation period. These secondary cases were excluded from any further risk factor analysis.

This study was conducted as a Public Health Investigation under Section 188 of the Victorian Public Health and Wellbeing Act 2008, and approval from a Human Research Ethics Committee (HREC) was not required. Contact with the control bank was encompassed by DHHS HREC approval of the population health survey in which they initially participated. Verbal consent was obtained prior to each interview, and if the case or control was under 18 years of age the consent of their parent or guardian was sought.

Environmental investigation

Public aquatic facilities are regulated in Victoria by the Public Health and Wellbeing Act 2008 and the Public Health and Wellbeing Regulations 2009. An aquatic facility outbreak was defined when 2 or more cases of cryptosporidiosis had swum at the same facility during their exposure period and case onset dates were less than 28 days apart. When linked cases were identified, facility information was referred to the DHHS Water Program and to local government environmental health officers for investigation and control activities. Control activities were guided by the DHHS Cryptosporidiosis Prevention and Outbreak Response Plan and the Pool Operators’ Handbook. To ensure any Cryptosporidium oocysts present in the aquatic facility were inactivated, hyperchlorination was requested. Water sampling is not incorporated in Victoria’s outbreak protocol and response plan due to measurement challenges associated with detection of Cryptosporidium oocysts, which are often in low abundance in the aquatic environment.

Results

Epidemiological investigation

Descriptive epidemiology

There were 70 notifications of cryptosporidiosis from the region of interest received in the period 1 March to 23 April 2015, which was higher than the 5-year average (2010–2014) for the same
period (55 notifications). Thirty-nine interviews were completed. Case exclusion was due to: non-contact (n=10); privacy reasons (n=11); refusal to participate (n=3); and inability to recall onset date (n=7). An additional 9 cases were excluded post interview as they either had an onset date after 21 March 2015 or were identified as secondary cases. Thirty cases and 66 controls were recruited. There were several peaks of onset of illness for included cases between 17 February and 21 March 2015, indicating the potential for multiple sources of infection (Figure). The median duration of diarrhoea for 27 of 30 cases was 13 days (range: 3–31 days); 3 cases still had diarrhoea at the time of interview. Two children under 8 years of age were hospitalised as a result of their illness. Other than diarrhoea, the most commonly described symptoms for cases were abdominal pain (88.2%), lethargy (82.4%) and nausea (76.5%). The age group and sex distribution of cases and controls are included in Table 1. The median age of cases was 20 years (range: 2–44 years), with 47% of cases aged 12 years or younger. Among adults, cases were predominantly aged 30–39 years. Approximately two-thirds (62%) of cases and controls were female.

Analytical epidemiology

Of the risk factors for Cryptosporidium infection that were explored, illness was significantly associated with recreational water exposure, specifically among those who reported swimming or paddling at a waterpark, and also with prior contact with a household member with a (non-specified) gastrointestinal illness (Table 2). Recreational water exposure in a public or private spa was also significantly associated with illness, although only 13% (4/30) of cases reported this exposure. There was no association between illness and recreational water exposure at public or private swimming pools, paddling pools or natural bodies of water. Consumption of bottled water was also identified as a risk factor for illness.

None of the other food based or animal contact risk factors examined were significantly associated with illness.

In multivariable analysis exposure at a waterpark or a public or private spa remained associated with illness, after adjusting for bottled water consumption and ill household contacts (Table 2). Bottled water consumption remained associated with illness in multivariable models, as did person-to-person transmission through household contacts with prior diarrhoeal illness.

Environmental investigation

Under DHHS criteria, 2 Victorian outdoor waterparks were implicated as exposure sites for 9 of the 11 cases in the current study who indicated they swam at a waterpark. Park A was located within the region (4 linked cases) and Park B in an adjacent region (5 linked cases). An additional 5 cases were identified as having attended Park A in the same period as included cases; however, these were excluded from the study analysis as they were either unable to recall the onset date (n=3) or resided outside of the region of interest (n=2). All cases reported waterpark attendance in their exposure period. More specifically, attendance was focused during a single weekend period in February 2015 at each of the 2 facilities: either 14–15 February (Park A) or 21–22 February (Park B).

Table 1: Age group and sex distribution of cases and controls in a study of cryptosporidiosis in a metropolitan region of Melbourne, February and March 2015

<table>
<thead>
<tr>
<th>Age group (years)</th>
<th>5 or under</th>
<th>6 to 12</th>
<th>13 to 19</th>
<th>20 to 29</th>
<th>30 to 39</th>
<th>40 to 49</th>
<th>Total</th>
<th>Sex (% female)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cases</td>
<td>6</td>
<td>8</td>
<td>1</td>
<td>6</td>
<td>6</td>
<td>3</td>
<td>30</td>
<td>62</td>
</tr>
<tr>
<td>Controls</td>
<td>14</td>
<td>20</td>
<td>2</td>
<td>9</td>
<td>14</td>
<td>7</td>
<td>66</td>
<td>64</td>
</tr>
</tbody>
</table>
Table 2: Age-adjusted univariable analysis and multivariable logistic regression for significant risk factors associated with *cryptosporidiosis* infection in one metropolitan region of Melbourne, February and March 2015

<table>
<thead>
<tr>
<th>Risk factor (excluding known secondary cases)</th>
<th>Cases (total = 30)</th>
<th>Control (total = 66)</th>
<th>Univariable</th>
<th>Multivariable</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Exposed</td>
<td>Total</td>
<td>%</td>
<td>Exposed</td>
</tr>
<tr>
<td>Other household member with diarrhoeal illness</td>
<td>8</td>
<td>30</td>
<td>27</td>
<td>4</td>
</tr>
<tr>
<td>Recreational water exposure (any)</td>
<td>24</td>
<td>30</td>
<td>80</td>
<td>37</td>
</tr>
<tr>
<td>Waterpark (any)</td>
<td>11</td>
<td>30</td>
<td>37</td>
<td>1</td>
</tr>
<tr>
<td>Waterpark (Victoria)</td>
<td>10</td>
<td>30</td>
<td>33</td>
<td>1</td>
</tr>
<tr>
<td>Spa (public or private)</td>
<td>4</td>
<td>30</td>
<td>13</td>
<td>1</td>
</tr>
<tr>
<td>Public pool</td>
<td>11</td>
<td>30</td>
<td>37</td>
<td>23</td>
</tr>
<tr>
<td>Private pool</td>
<td>7</td>
<td>30</td>
<td>23</td>
<td>9</td>
</tr>
<tr>
<td>Natural bodies of water†</td>
<td>5</td>
<td>30</td>
<td>17</td>
<td>11</td>
</tr>
<tr>
<td>Paddling pool</td>
<td>0</td>
<td>30</td>
<td>0</td>
<td>8</td>
</tr>
<tr>
<td>Drank bottled water</td>
<td>24</td>
<td>28</td>
<td>86</td>
<td>35</td>
</tr>
</tbody>
</table>

OR: odds ratio; 95% CI: 95% confidence interval.

* Mantel-Haenszel OR adjusted for matched age-group.
† OR adjusted for all other variables in model, including age-group.
‡ Natural bodies of water include river, lake, dam, bore water or beach.
Local government environmental health staff liaised with the facility manager at Park A and identified deficient filtration systems in several pools, including a shallow play area with water fountains, splash slides and spray areas. Hyperchlorination of all pools at the Park A was conducted to inactivate any Cryptosporidium oocysts present in the water, during which time the park was closed to patrons. The local government environment health unit in the region where Park B was located had been advised by members of the public and other council staff of anecdotal cases of gastrointestinal illness in large groups following attendance over a single weekend. As a result, Park B undertook voluntary hyperchlorination prior to DHHS identifying that notified cases were linked to this facility. In addition, DHHS also provided information to staff at both facilities for distribution to patrons about preventative healthy swimming practices, including diarrhoea exclusion periods, personal swim hygiene, use of designated nappy change areas, and avoiding swallowing pool water.

**Discussion**

The increase in cryptosporidiosis notifications observed in an urban region of Melbourne, Victoria, in February and March 2015 was strongly associated with recreational water exposure, specifically at waterparks. In addition, cases were temporally linked with attendance at 2 waterpark facilities, one of which had evidence of deficiencies in water quality maintenance. This investigation resulted in the execution of immediate control activities at these sites. Internationally, outbreaks of cryptosporidiosis associated with water or splash parks have been reported in the United States of America, Canada and the United Kingdom. Our investigation highlights that waterparks also require specific attention for the prevention of cryptosporidiosis outbreaks in Australia.

_Cryptosporidium_ are well suited to transmission in recreational water, and outbreaks can occur in aquatic facilities even if they are well maintained, as swimmers can be exposed to _Cryptosporidium_ oocysts during the time it takes for water to be disinfected or filtered. As such, control of transmission relies on both good swimmer hygiene to prevent the introduction of oocysts into recreational waters, and effective pool engineering controls for water filtration and treatment.

The design and nature of use of waterparks, which can differ from standard pool facilities, raise unique considerations for directing prevention and control activities. Waterparks, including those identified in the current investigation, typically include water play areas such as splash zones, fountains and shallow wading pools: features which have been suggested to contribute to the transmission of infection in other studies. While splash activities may lead to increased swallowing of water, increased use of shallow pools by children wearing nappies, sitting on splash features and accidental faecal release incidents are also suggested to contribute to _Cryptosporidium_ infection in toddler pools at waterparks. Although pools frequented by toddlers are the most plausible source of contamination, a specific wading pool or splash activity was not implicated at either facility in this study.

Responding to an increased number of waterpark-related outbreaks, the Centers for Disease Control and Prevention (CDC) in the United States of America identified that specific patron messaging needed to be designed for water play and interactive areas, to prevent contamination and reduce potential for transmission. It is recommended that similar messaging be developed for use in Australia in addition to existing healthy swimming advice.

Outdoor shallow toddler pools and splash pools present at waterparks inherit special problems associated with their engineering and maintenance. In addition to contamination challenges in toddler pools outlined above, outdoor pool water quality can be compromised through chlorine degradation by UV light and their potential to be polluted by other foreign matter that blows into them. Waterparks can be popular destinations in peak periods. Evidence from this study and other investigations suggests that individuals often travel a greater distance to attend these specialised recreational water facilities. As such, high bather densities can further challenge pool maintenance, especially in shallow toddler pools or splash pools where bather pollution can be high for the amount of water involved.

It has been highlighted elsewhere that waterpark operators cannot solely rely on filtration and chlorine disinfection to protect patrons from _Cryptosporidium_. In alignment with other advice, in order to minimise transmission of _Cryptosporidium_ infection, we also recommend that separate maintenance systems should be applied to pools with high infant bather loads. These may encompass the use of separate backwash and filtration systems, regular filter maintenance and hyperchlorination procedures, and more frequent pool content circulation.

In this study, almost half of the cases were aged 12 years or younger and another third were adults aged 30–39 years. This pattern is consistent with the bimodal age distribution of _Cryptosporidium_.
notification rates in Victoria observed between 2001 and 2009, while children are suggested to be more susceptible to parasitic infections, other researchers have noted that this distribution may also reflect notification bias, with medical care more likely to be sought for children. Additionally, increased swallowing of water when swimming or playing in shallow pool areas and splash zones (such as those identified in the implicated waterparks) may mean that children have a greater risk of being infected.

Interestingly, after adjusting for other confounding factors, consumption of bottled water remained significantly associated with illness. This finding is in contrast to those of a study of a large statewide outbreak of cryptosporidiosis in New South Wales in 1997 to 1998, which found that cases were less likely to report drinking bottled water. It is feasible that bottled water is a potential source of cryptosporidiosis as other international studies have identified Cryptosporidium oocysts in commercially bottled water and a case-control study conducted in South Australia in 1993 identified consumption of bottled spring water as risk factor for illness.

In the current study, information wasn’t specifically collected on brands or amounts of bottled water drunk by cases and controls to enable further examination of this association. Furthermore, the association of illness with bottled water consumption in this study may have limited plausibility given that we know from the department’s recent food frequency survey (Marion Easton, personal communication) that around 33% of Victorians surveyed, regardless of the time of year, drank bottled water. It is likely then, that if there was a true association between drinking bottled water with illness it would have been expected that the impact of this would have been observed Victoria-wide, rather than limited to a specific region. It is also possible that this association may be a result of uncontrolled confounding, as several studies have described sociodemographic factors that are associated with bottled water use through their influence on consumer perception of water risk and quality.

To a lesser extent, the odds of illness were also increased among those cases who used public or private spas and person-to-person transmission in a household environment. Although not significantly associated with the rise in cases in the region, interviews with cases also highlighted the diversity of risk factors associated with sporadic Cryptosporidium infection, which otherwise contributed to background rates in this region and would have also been captured in the study population.

These findings need to be considered within the limitations of this study. As controls were not contacted until after all cases were interviewed, and were asked about activities over a similar time period as cases, this may have resulted in a systematic difference in recall in controls. Furthermore, case exclusions may have meant that not all risk factors for cryptosporidiosis were fully elucidated.

Outbreaks of cryptosporidiosis can be difficult to identify, as it can often be a mild, self-limiting illness for which medical attention is not sought. Despite strong epidemiological evidence, this study did not include a microbiological investigation for evidence of water contamination at the implicated facilities. It may be that species identification and molecular sub-typing of Cryptosporidium may have strengthened links between cases and exposure sites and also provided further evidence for the diversity of sources of infection; however, a number of studies exist that provide a rich body of evidence in this area.

While sub-typing may have assisted in the attribution of causality, Cryptosporidium species differentiation is not routinely performed in Australian diagnostic laboratories and cannot currently be used for retrospective public health investigations in Victoria. An enhanced environmental investigation at each facility may have served to understand individual and overall pool treatment and backwash systems and identify specific deficits in these, as well as the incidence and management of faecal incidents.

Conclusion

This study has shown that recreational water facilities, including waterparks, remain high risk sites for the acquisition of Cryptosporidium infection. This assertion is further strengthened by the identification of cases linked to attendance at 2 separate waterparks during this investigation.

The maintenance of water quality at aquatic facilities requires vigilance by both operators and the public. This study is an important reminder of the need for preventive risk management through healthy swimming practices and ensuring the adequate performance of treatment processes within aquatic facilities. Specific recommendations include the development of healthy splash advice specific for waterpark patrons, and the use of separate backwash and filtration systems for high risk pools and activities.
Author details
Dr Tanyth E de Gooyer, Master of Philosophy (Appl. Epi) Scholar
Ms Joy Gregory, Principal OzFoodNet Epidemiologist
Ms Marion Easton, OzFoodNet Epidemiologist
Dr Nicola Stephens, Manager, Communicable Disease Epidemiology and Surveillance
Dr Emily Fearnley, Research Fellow
Associate Prof Martyn Kirk, Convener, Master of Philosophy in Applied Epidemiology

1. Victorian Department of Health and Human Services, Melbourne, Victoria
2. National Centre for Epidemiology and Population Health, Australian National University, Acton, Australian Capital Territory

Corresponding author: Dr Tanyth de Gooyer, Department of Health and Human Services, 50 Lonsdale Street, MELBOURNE VIC 3001. Telephone: +61 3 9096 5308. Email: tanyth.degooyer@dhhs.vic.gov.au

References
28. Franco RM, Cantusio Neto R. Occurrence of crypto-
sporidial oocysts and giardia cysts in bottled min-
eral water commercialized in the city of Campinas,
State of Sao Paulo, Brazil. *Mem Inst Oswaldo Cruz*

29. Weinstein P, Macaitis M, Walker C, Cameron S.
Cryptosporidial diarrhoea in South Australia. An explor-
atory case-control study of risk factors for transmission.

30. Hu Z, Morton LW, Mahler RL. Bottled water: United
States consumers and their perceptions of water quality.

31. Robertson B, Sinclair MI, Forbes AB, Veitch M, Kirk M,
Cunliffe D, et al. Case-control studies of sporadic
cryptosporidiosis in Melbourne and Adelaide, Australia.

32. Hunter PR, Hughes S, Woodhouse S, Syed Q,
Verlander NQ, Chalmers RM, et al. Sporadic crypto-
sporidiosis case-control study with genotyping.

33. Chalmers RM, Smith R, Elwin K, Clifton-Hadley FA,
Giles M. Epidemiology of anthroponotic and zoonotic
human cryptosporidiosis in England and Wales, 2004–

34. Roy SL, DeLong SM, Stenzel SA, Shiferaw B, Roberts JM,
Khalakdina A, et al. Risk factors for sporadic crypto-
sporidiosis among immunocompetent persons in the
2004;42(7):2944–2951.

35. Ng J, Eastwood K, Durrheim D, Massey P, Walker B,
Armson A, et al. Evidence supporting zoonotic transmis-
sion of Cryptosporidium in rural New South Wales.