DETECTION OF CRYPTOSPORIDIUM SP. OOCYSTS IN GROUNDWATER FOR HUMAN CONSUMPTION IN ITAQUAQUECETUBA CITY, S. PAULO-BRAZIL

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Submitted: October 29, 1998; Returned to authors for corrections: October 08, 1999; Approved: June 26, 2000

SHORT COMMUNICATION

ABSTRACT

Cryptosporidium is an emergent pathogen that causes profuse diarrhea in humans. Outbreaks of human cryptosporidiosis have implicated water as a possible source of contamination. In this study the presence of Cryptosporidium sp. oocysts was investigated as well as the fecal contamination in groundwater for consumption in Itaquaquecetuba – São Paulo. Therefore, it was possible to notice that the septic tanks were located close to the wells, exposing them to risk of contamination. As a consequence of these results and observations efficient disinfecting practices and groundwater monitoring are recommended.

Key words: Cryptosporidium sp. oocysts, calcium carbonate flocculation method, and groundwater

Cryptosporidium is a protozoan parasite that causes profuse diarrhea in humans. Cryptosporidiosis can be transmitted by fecal-oral route. Recent reports have suggested an association between cryptosporidiosis and water supplies (10). The presence of Cryptosporidium sp. oocysts in water supply sources constitutes a potential risk of waterborne cryptosporidiosis outbreaks and has been a matter of concern for public health departments and public water system authorities.

Cryptosporidium sp. oocysts are highly resistant to chemical disinfectants such as chlorine used to treat drinking water. Physical removal of the parasite from water by filtration is an important process in water treatment (8, 11). Studies showed that ozone, high temperature or chlorine dioxide are effective in removing or inactivating oocysts (4, 5).

In Brazil, according to the Ministry of Health (2), 2842 cases of cryptosporidiosis were reported in AIDS patients, during the period 1980 – 1997. Sauda (12) confirmed the prevalence of oocysts in AIDS patients at Santos City, São Paulo (SP) State Brazil. Infections caused by Cryptosporidium were also reported in impoverished communities of Fortaleza, Ceará State Brazil. In this city, local contamination of water supplies and infection of household animals contribute to the maintenance of this infection (9, 16). Tomps (14) reported Cryptosporidium sp. oocysts occurrence in fecal and water samples in Perus District – SP. CETESB (Companhia de Tecnologia de Saneamento Ambiental) the environmental protection agency of São Paulo, has also found positive samples in shallow wells in Mauá District - SP.

One of the problems for surveillance of the disease is the difficulty in detecting the pathogen in environmental samples. Therefore, different methods have been developed in order to detect this protozoan in water samples (13). Environmental Protection Agency (3) proposed some of them, but there are not standardized techniques yet.

Our previous data (not shown) were undertaken to investigate the efficiency of the concentration methods by filtration (3) and calcium carbonate flocculation method (15). This last method demonstrated to be simple and economical, due to the need of
common equipment and considerably less labor than other ordinary methods.

Taken the potential risk of this emergent pathogen and the importance of data about its prevalence in Brazil, the goal of this study was to investigate the presence of Cryptosporidium sp. oocysts and the fecal contamination in groundwater used for consumption in Itaquaquecetuba City – SP (Fig. 1A), an area showing high infantile mortality rate. Most of the households have poor sanitary facilities and no sewer line. The community is supplied by groundwater from shallow wells and the domestic sewage goes to septic tanks. At least one animal dwells either the house or the backyard. The vast majority are pet animals (dogs and cats), although chickens, ducks, pigs and cows are found as well.

Groundwater samples (10L) were collected from 10 shallow wells and 2 samples from septic tanks located in regions considered risk areas either due to their proximity to a septic tank/well or to inadequate construction (Fig. 1B). The samples were concentrated and processed using a carbonate flocculation method (15). The direct immunofluorescence assay (IFA MERIFLUOR – MERIDIAN. Diagnostics, Inc.) was used to check the pathogen presence (7).

The evidence of fecal contamination in wells was verified through detection of E.coli by Multiple-Tube Technique (1) using chromogenic and fluorogenic substrate (Colilert 18, IDDEX).

The presence of Cryptosporidium sp. oocysts was detected in 8 shallow wells and in the 2 septic tanks samples (Table 1). The positive results obtained from septic tanks suggest possible risk for contamination of groundwater. According to the World Health Organization the nearness of septic tanks and wells is the major cause of infant mortality (6). The result support that there is a potential risk of waterborne disease to the community, once they consume the water, without boiling it.

The results suggested that the presence of Cryptosporidium sp. oocysts in groundwater samples are not associated with the presence of E. coli, in agreement with the literature (13) (Table 1). New bacteriological indicators should be taken into account in order to confirm the absence of these persistent pathogens.

In Brazil, groundwater is an important natural resource and must be protected from different sources of contamination. Efficient disinfecting practices and monitoring for the presence of Cryptosporidium sp oocysts is strongly recommended for water sources in the city of São Paulo.

Table 1. Results of detection of E.coli and Cryptosporidium sp oocysts in shallow wells and septic tank samples

<table>
<thead>
<tr>
<th>Samples</th>
<th>E. coli (MPN/100 ml)</th>
<th>Cryptosporidium sp. oocysts</th>
</tr>
</thead>
<tbody>
<tr>
<td>W04</td>
<td>2</td>
<td>P</td>
</tr>
<tr>
<td>W19</td>
<td>170</td>
<td>P</td>
</tr>
<tr>
<td>W20</td>
<td>&lt;2</td>
<td>A</td>
</tr>
<tr>
<td>W21</td>
<td>220</td>
<td>P</td>
</tr>
<tr>
<td>W26</td>
<td>&lt;2</td>
<td>A</td>
</tr>
<tr>
<td>W29</td>
<td>130</td>
<td>P</td>
</tr>
<tr>
<td>W42</td>
<td>2</td>
<td>P</td>
</tr>
<tr>
<td>W44</td>
<td>110</td>
<td>P</td>
</tr>
<tr>
<td>W45</td>
<td>2</td>
<td>P</td>
</tr>
<tr>
<td>W46</td>
<td>&lt;2</td>
<td>P</td>
</tr>
<tr>
<td>Septic tanks</td>
<td>S 01</td>
<td>≥1600</td>
</tr>
<tr>
<td></td>
<td>S 02</td>
<td>≥1600</td>
</tr>
</tbody>
</table>

W – wells
S – septic tanks
P – presence
A – absence
MPN – most probable number

ACKNOWLEDGEMENTS

This study was supported by FAPESP proc. nº 1996/12756-5.

RESUMO

Detecção de oocistos de Cryptosporidium sp. em águas de poço para consumo humano em Itaquaquecetuba, região metropolitana de São Paulo

Cryptosporidium é um patógeno emergente que causa diarreia em humanos. Muitos surtos graves de criptosporidiase tem a água como possível origem da contaminação. Neste estudo foi investigada a presença do oocisto de Cryptosporidium sp.
em águas de poço, usadas para consumo humano em Itaquaquecetuba - São Paulo. Os resultados evidenciaram a presença de oocistos em 2 fossas e em 8 poços analisados, mostrando o risco que esta água representa para consumo humano e a necessidade de práticas mais eficientes de desinfecção e monitoramento da presença deste parasita em amostras de águas no estado de São Paulo.

**Palavras-chave:** oocistos de *Cryptosporidium*, floculação com carbonato de cálcio, águas subterrâneas.

### REFERENCES