

A serological study of *Cryptosporidium* transmission in a periurban area of a Brazilian Northeastern city

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Summary

OBJECTIVES To study the prevalence of *Cryptosporidium* infection by measuring the levels of anti-*Cryptosporidium* IgG antibodies among people inhabiting three neighbourhoods of a periurban area of Salvador, Northeast of Brazil; and to investigate the effects of environmental sanitation measures, hygienic habits and household water supply, storage and handling on the frequency of these antibodies in sera of the studied population.

METHODS *Cryptosporidium* inter-household transmission was studied by comparing the frequency of anti-*Cryptosporidium* IgG antibodies among people inhabiting areas with or without different environmental sanitation measures and intra-household transmission by comparing the presence of these antibodies in families with or without cases of diarrhoea, associated with the presence of *Cryptosporidium* oocysts in their stools. Children or family members with diarrhoeal episodes were evaluated parasitologically for *Cryptosporidium* infection by testing stool specimens with the Ritchie-modified formol-ether concentration and the acid-fast staining methods. All groups were serologically evaluated for parasite exposure by an indirect enzyme-linked immunosorbent assay.

RESULTS A statistically significant difference was detected in the prevalence of *Cryptosporidium* infection between area 1 which had no environmental sanitation measures and area 3 which had improved environmental sanitation measures ($P = 0.044$). Most of the hygienic habits investigated did not correlate with the presence of anti-*Cryptosporidium* antibody in sera of the population studied. However, positive associations were found between both poor household water supply (OD = 0.17; 90% CI = 0.09–0.32; $P = 0.0001$) and drinking unboiled/unfiltered water (OD = 0.40; 90% CI = 0.24–0.67; $P = 0.0002$) with high levels of anti-*Cryptosporidium* antibodies in sera.

CONCLUSIONS These data suggest that although uncorrected household water supply, storage and handling play an important role on *Cryptosporidium* transmission in periurban areas of developing country cities, like Salvador, Brazil, inadequate environmental conditions may also contribute to the spread of this parasite.

keywords *Cryptosporidium*, seroprevalence, transmission, environmental sanitation measures, water supply

Introduction

Cryptosporidium species are widespread protozoa and largely detected in environmental samples which infect both humans and animals. The parasite can cause acute and persistent disease in malnourished children and chronic, life-threatening infection in immunodeficient individuals, such as those with AIDS (O'Donoghue 1995; Ramirez *et al.* 2004). In healthy subjects living in developed

countries, cryptosporidiosis is acute and short-term, mostly epidemically (D'Antonio *et al.* 1985; MacKenzie *et al.* 1995), except in cases of traveller's diarrhoea, nosocomial or nursery outbreaks (CDC 1984; Goodgame *et al.* 1995). The prevalence of cryptosporidiosis during non-epidemic periods is low, even in immunosuppressed individuals (Sorvillo *et al.* 1998). In contrast, outbreaks of cryptosporidiosis among healthy adults are less frequently reported in developing countries, although this disease is

found in 6–41% of children and in 14–48% of AIDS patients with diarrhoea illnesses (Guerrant *et al.* 1990; Guerrant 1997).

Contamination of water supplies by animal or human faeces containing *Cryptosporidium* *sp* oocysts seems to play a major role in the causality of the cryptosporidiosis epidemics reported in developed countries (D'Antonio *et al.* 1985; MacKenzie *et al.* 1995; Hlavsa *et al.* 2005), while the transmission mechanisms of these parasites in low income population have not yet been completely uncovered. As many inhabitants of periurban areas in developing countries live in an environment without adequate sanitation measures and with poor inter- and intra-household hygiene practices, with propitious conditions for the survival and propagation of *Cryptosporidium* oocysts, the role of water contamination in these environments may be less evident in the transmission of this parasite.

Laboratory methods to confirm *Cryptosporidium* infection are hampered by the large number of oocysts per gram of stool needed (Weber *et al.* 1991). Evaluation of exposure to the parasite by serology is much more sensitive than detection of oocysts in stool samples, especially in populations chronically exposed to *Cryptosporidium* through contaminated food or drinking water, and has been widely used for epidemiological studies (Kuhls *et al.* 1994; Zu *et al.* 1994).

In our effort to study inter-household transmission, we compared the levels of anti-*Cryptosporidium* IgG antibodies among people inhabiting an area without environmental sanitation measures with two areas with different environmental sanitation measures in place. Intra-household transmission was studied by comparing anti-*Cryptosporidium* IgG antibody levels in families with or without cases of diarrhoea associated with presence of *Cryptosporidium* oocysts in their stools. Finally, we investigated the effects of the same environmental sanitation measures and inter- and intra-household water supply, storage and handling on the levels of anti-*Cryptosporidium* IgG antibodies in sera of the studied population.

Materials and methods

Population and environments studied

The population studied consisted of 138 children aged 0–4 years followed-up for a diarrhoea diseases study (Morales 1996) and 235 relatives of these children, living in areas with the following levels of environmental sanitation facilities: area 3 with stairways and a ramps drainage system for collecting and transporting storm

water plus a simplified sewerage system for collecting and transporting sewage; area 2 with only stairways and a ramps drainage system for collecting and transporting storm water and sewage; area 1 without interventions. In area 1, human excreta and sewage were either disposed off in a backyard or in an open sewer in a public environment. The children had their stools examined for *Cryptosporidium* oocysts in diarrhoeal episodes and, at the end of follow-up, all children, along with the members of their families, had a blood sample taken to determine their anti-*Cryptosporidium* IgG antibody levels. Families of children who had diarrhoeal episodes and *Cryptosporidium* oocysts identified in their stools (case families) were compared with families of children with diarrhoeal episodes who tested negative for *Cryptosporidium* oocysts shedding (control families). Child relatives were interviewed for history of diarrhoea, water supply, storage and handling, environmental and household sanitary conditions. Variables on water supply (treated water supplied by the city council supplier or another source), storage (open/closed containers or other vessels), use (all uses, drinking, cooking and personal hygiene), handling in the house (if boiled or filtered) and frequency of water supply distribution (regular or not) were investigated.

Likewise, some variables on household sanitation facilities were investigated, such as presence of excreta and solid waste disposal facilities, toilette type (flush, pit latrine, none), presence of washstand, public removal and frequency of rubbish collection.

In this study, inter-household transmission was studied by comparing the prevalence of anti-*Cryptosporidium* IgG serum antibodies among the population living in areas with different environmental sanitation measures. It was thought that parasite transmission might occur in an indirect fashion – in the surrounding environment frequented by youngsters for long periods of time, in contact with contaminated soil. Intra-household transmission was studied by comparing the presence of anti-*Cryptosporidium* antibodies between individuals dwelling with, or not dwelling with, children excreting *Cryptosporidium* oocysts in their stools, which was detected by fecal examination. In this case, the transmission of oocysts could be either person-to-person or indirectly, i.e. by ingestion of food/water contaminated with faeces, or even during contact with the house soil.

Medical procedures of this work were approved by the Medical Research Ethical Committee of the Hospital Professor Edgar Santos, Federal University of Bahia. All adult participants signed a form stating that they and their family members joined the studied group voluntarily.

Stool examination

Fresh stool specimens were sieved to remove coarse fibres and submitted to the Ritchie-modified formol-ether concentration method (Ritchie 1948). The pellet from each sample was spread on two glass slides, dried and stained by the acid-fast staining (Henriksen & Pohlenz 1981).

Detection of anti-*Cryptosporidium* IgG antibody in serum samples

To detect *Cryptosporidium* antibodies in serum samples, an indirect enzyme-linked immunosorbent assay (ELISA) was performed, using oocyst lysate as antigen. *Cryptosporidium* oocysts were obtained from faeces of an experimentally infected-calf and purified by discontinuous sucrose and Percoll gradients (Arrowood & Sterling 1987). Wells of microassay plates were incubated with 1 µg of crude *Cryptosporidium* antigen in 100 µl of carbonate buffer, pH 9.5, overnight at 4 °C. After blocking with 1% casein (Sigma Chemicals, St Louis, MO, USA) in 0.15 M sodium phosphate-buffered saline, pH 7.2 (PBS), the wells were incubated with a 1:125 dilution of the test sera. After washings, anti-human IgG peroxidase-conjugated (Sigma Chemicals) was diluted at 1:2000 and was applied. All washings and sera and conjugate dilutions were done with PBS containing 0.5% casein and 0.05% Tween 20 (Sigma Chemicals), and all incubations were performed for 1 h at 37 °C. Finally the plates were washed and the substrate containing tetramethylbenzidine and hydrogen peroxide (Sigma Chemicals) was applied. The reaction was terminated after 15 min with the addition of 2 N sulphuric acid to the wells and subsequently read at 450 nm filter using a microtiter reader (Dynatech Laboratories, Alexandria, VA, USA). The assay cut-off was obtained as the mean plus two standard deviations of sera from 10 children of high socioeconomic level, without any diarrhoea episodes and with several faecal samples submitted to *Cryptosporidium* oocysts identification by the modified acid-fast staining technique.

Statistical analysis

The epidemiological data were analysed with the aid of the EPI-INFO and SPSS Packages for Windows (SPSS 2001). The Pearson chi-square test was used to verify independent variables. The test for proportions was used to verify differences among age groups, areas of study and presence of *Cryptosporidium* antibodies. The odds ratio (OR) used to analyse sanitation and water management measures and the presence of *Cryptosporidium* antibodies in the

individuals' sera had its significance level obtained by the chi-square test. $P < 0.05$ was considered significant.

Results

Cryptosporidium IgG levels in sera of the studied population by gender, age and level of sanitation facilities available in the living areas

The overall prevalence of *Cryptosporidium* antibodies in the sera of 332 individuals assessed by indirect ELISA was 57.5% (Table 1); 152 individuals were male (45.8%) and 180 (54.2%) were female; no statistically significant differences were found between gender and presence of *Cryptosporidium* antibodies in the sera of these individuals ($P = 0.322$; Pearson chi-square). 138 (41.6%) individuals were 0–4 years old; 103 (31.0%) 5–20; 79 (21.4%) were aged 21–35 years and 20 (6.0%) were older than 35 years. The chi-square for comparisons of proportions did not show statistically significant differences among these age groups ($P = 0.919$) and humoral response to *Cryptosporidium*. One hundred and thirty-two individuals (39.8%) lived in area 1, without environmental sanitation interventions; 134 (40.4%) lived in area 2, with stairways and a ramps drainage system collecting both storm water and sewage and 66 (19.9%) lived in area 3, where stairways, ramped drainage system and simplified sewerage systems were distinct. When analysed together, the differences among living in any of the three areas and the presence of *Cryptosporidium* antibodies in the sera of the inhabitants were not statistically significant ($P = 0.076$; chi-square for comparisons of proportions). However, a difference was found when area 1 was compared with area 2 ($P = 0.064$) and became statistically significant when area 1 was compared with area 3 ($P = 0.044$).

Cryptosporidium antibody levels in sera of members of families with and without a case of cryptosporidiosis

A total of 709 faecal samples from the children were examined during diarrhoeal episodes, and *Cryptosporidium* oocysts were found in 4.2% of the samples. *Cryptosporidium* IgG was assayed in 55 sera from the relatives of 14 children who had at least one diarrhoeal episode with *Cryptosporidium* oocysts detected in their faeces (case families) and in 119 sera from relatives of 28 children followed up, who never had *Cryptosporidium* oocysts detected in their faeces (control families; Table 2). Three-quarters (74.5%) of the individuals from case families and 62.1% of the individuals from control families were positive for *Cryptosporidium* antibodies (OR = 1.91; CI = 0.89–4.14; $P = 0.07$).

Table 1 Seroprevalence of anti-*Cryptosporidium* IgG antibodies by age, sex and living area of the studied population

Demographic data	<i>n</i>	%	ELISA positivity*		Statistic tests (90% CI) (chi-square for comparison of proportions)
			<i>n</i>	%	
Age (years)					
0–4	138	41.6	82	59.4	<i>P</i> = 0.919 (in all groups)
5–20	103	31.0	57	55.3	
21–35	71	21.4	40	56.3	
>35	20	6.0	12	60.0	
Total	332	100	191	57.5	
Sex (Pearson chi-square)					
Male	152	45.8	83	54.6	<i>P</i> = 0.322
Female	180	54.2	108	60.0	
Total	332	100	191	57.5	
Living area					
1	132	39.8	86	65.1	<i>P</i> = 0.064 (area 1 and 2)
2	134	40.4	71	52.9	
3	66	19.9	34	51.5	<i>P</i> = 0.044 (area 1 and 3)
Total	332	100	191	57.5	<i>P</i> = 0.076 (in all areas)

*The prevalence of anti-*Cryptosporidium* IgG antibodies was assessed by enzyme immunoassay (ELISA), as described in Materials and methods, in sera of individuals living in areas with the following sanitation measures: (1) without intervention; (2) stairways and ramps drainage system collecting and transporting storm water and sewage; (3) stairways and ramps drainage system and simplified sewerage system distinct.

Table 2 Prevalence of anti-*Cryptosporidium* antibody in individuals from families with or without a case of cryptosporidiosis

Families	ELISA*		Total <i>n</i> (%)	Odds ratio	CI (90%)	<i>P</i> -value
	Negative <i>n</i> (%)	Positive <i>n</i> (%)				
Case†	14 (25.5)	41 (74.5)	55 (100)	1.91	0.89–4.14	0.07
Control‡	47 (37.9)	72 (62.1)	119 (100)			

*Anti-*Cryptosporidium* IgG antibodies were detected in individuals from families with (†) or without (‡) a case of cryptosporidiosis in children, detected by the presence of oocysts in faecal samples by the modified Ziehl-Neelsen technique as stated in Materials and methods.

Cryptosporidium infection in a periurban area of Salvador, Brazil is associated with poor household and environmental sanitation conditions

The following household and environmental sanitation measures were not statistically found to be associated with the presence of *Cryptosporidium* antibodies in the sera of the surveyed population: presence of flush toilet in the household; presence of a washstand; adults and children sharing the same flush toilet; rubbish storage at home or disposed of in the street, or frequency of rubbish collection (data not shown); drinking water distinct from cooking water (OR = 0.53; *P* = 0.065) and household in-door water tubing/plumbing (OR = 0.68–2.23; *P* = 0.2) (Table 3). However, some household and environmental sanitation measures were statistically associated with the presence of *Cryptosporidium* antibody in the sera of the

studied population. These measures could be grouped as positive (related to reducing antibody frequency) such as the systematic cleaning of stairways and ramped drainage systems by SUMAC (the public storm water maintenance service) (OR = 0.47; *P* = 0.017); drinking water provided by EMBASA (the Water Supply and Sanitation Public Company of the State of Bahia that distributes treated water; OR = 0.17; *P* = 0.000001); storage of drinking water in closed containers (OR = 0.44; *P* = 0.02); and water boiling/filtering before drinking (OR = 0.4; *P* = 0.0002); or as negative (related to increasing antibodies frequency), as water supply distribution shortage (OR = 1.95; *P* = 0.041); (Table 3). Unfortunately, one shortcoming of our study was that we did not evaluate hand-washing habits, although the presence of a washstand in households may reflect the influence of this hygiene habit in *Cryptosporidium* infection.

Table 3 Relationship between water supply, storage and handling and anti-*Cryptosporidium* IgG antibodies

Variable	Presence	ELISA positivity*		n	Odds ratio	CI (90%)	P-value
		n	%				
Water supply comes from EMBASA†	No	84	83.1	101	0.17	(0.09–0.32)	0.0001
	Yes	107	47.3	231			
Water supply shortage	No	19	43.1	44	1.95	(0.97–3.95)	0.041
	Yes	145	59.6	243			
Household in-door water pipe	No	44	24.1	182	1.24	(0.68–2.23)	0.2
	Yes	28	26.6	105			
Keep water in household in closed containers	No	130	62.8	207	0.44	(0.25–0.76)	0.020
	Yes	34	42.5	80			
Drinking water differ from cooking water	No	147	59.2	248	0.53	(0.26–1.11)	0.065
	Yes	17	43.6	39			
Boil or filter drinking water	No	102	67.5	151	0.40	(0.24–0.67)	0.0002
	Yes	62	45.8	136			
SUMAC cleans the stairways and ramps drainage system‡	No	144	60.3	239	0.47	(0.24–0.92)	0.017
	Yes	20	41.6	48			

*Anti-*Cryptosporidium* antibody was detected in sera from individual which neighbourhoods and households were surveyed for the different environmental sanitation facilities.

†The Water Supply and Sanitation Public Company of the State of Bahia.

‡City council storm water system maintenance provider.

Discussion

The overall 57.5% seroprevalence of *Cryptosporidium* IgG antibodies we found is close to that described in two low socioeconomic populations of Peru and Venezuela (Ungar *et al.* 1986) and relatively high considering the low prevalence of parasitologically confirmed cases among the studied population. However, it is much lower than that reported in children from a slum neighbourhood in Fortaleza, Brazil (Newman *et al.* 1994). Seroprevalence of cryptosporidiosis may vary from 46% to 89% in populations that inhabit the same city or state, depending on the level of exposure to parasite infection risk factors, such as source and type of treatment of drinking water, diet, food hygiene, household income, animal exposure and environmental conditions (Leach *et al.* 2000; McDonald *et al.* 2001; Frost *et al.* 2005). Differences in *Cryptosporidium* exposure risk factors (children from slums in the city of Fortaleza may live at higher risk than those from Salvador), combined with a variation in ELISA procedures, such as serum dilution, may contribute to the inter-study differences in *Cryptosporidium* seroprevalences among Brazilian children. Even in our study these differences could be observed between the whole population and the case-control groups seroprevalences, being higher in the latter one. This finding was attributed to the fact that most of the index cases used to select the family case and their controls came from area 1, with no environmental sanitation measures (data not shown).

Cryptosporidium antibodies among relatives of children shedding (74.5%) or not (62.1%) oocysts, although higher in the former group, did not show statistically significant differences. We credited the failure in demonstrating a statistically significant difference between these two groups to the small numbers of individuals in both groups. It is possible that increasing these studied groups would more efficiently show this potential difference.

The elucidation of the *Cryptosporidium* transmissibility mechanism may help the adoption of control measures for interrupting the parasite spreading in population. Major sources of infection include oocyst-contaminated water or food by animal/human faeces and household contact with infected persons (Leach *et al.* 2000; Ramirez *et al.* 2004). We found a positive association between the presence of *Cryptosporidium* antibody in population sera and poor practices of water management, as described previously (D'Antonio *et al.* 1985; Atherton *et al.* 1995; MacKenzie *et al.* 1995). In cities in the developed world, occasional disturbances of water supply systems, such as contamination by animal manure or breaking of filtering systems are responsible for epidemics, which occur in previously non-exposed populations (D'Antonio *et al.* 1985; MacKenzie *et al.* 1995; Fayer 2004). On the other hand, in developing country cities, closer proximity of human faeces to water sources may promote these disturbances more frequently. Furthermore, not all populations have access to the public water supply system.

In this work, the measures associated either with protection from *Cryptosporidium* infection, or with susceptibility to this parasite, were mostly related to the type or water supply management. This could be specifically observed by the high statistically significant difference between the prevalence of *Cryptosporidium* antibodies in the sera of individuals with and without access to treated water from EMBASA, and between individuals whose homes had or did not have a shortage in the distribution of this water supply service. Furthermore, contamination of drinking water by sewage or the use of sewage sludge in agriculture can be a source of population infection by several waterborne protozoa including *Cryptosporidium*. In fact, studies in Brazil have reported the occurrence of *Cryptosporidium* oocysts in: (1) sewage samples (Iacovski *et al.* 2004; Santos *et al.* 2004); (2) water samples from watersheds (Hachich *et al.* 2004) or from urban storm water systems (Newman *et al.* 1993); and (3) superficial raw water from a river (Franco *et al.* 2001). As *Cryptosporidium* oocysts are highly resistant to chemical disinfectants, the physical removal of oocysts by filtration is very important in treatment of water supply sources (Carey *et al.* 2004). Indeed, the resistance of oocysts to chlorine treatment of drinking water and the inefficiency of coarse filtration methods to remove this very small parasite are the main factors contributing to cryptosporidiosis outbreaks in developed countries (D'Antonio *et al.* 1985; Atherton *et al.* 1995; MacKenzie *et al.* 1995).

The human immune response against *Cryptosporidium*, although not long lasting, is protective. Therefore, the presence of antibodies may protect the immunocompetent individual from severe forms of cryptosporidiosis and many asymptomatic infections may occur in these populations (Campbell & Current 1983; Riggs 2002). Alternatively, these antibodies might only be markers of another protective immune response against this parasite. Indeed, high prevalence rates of clinically manifested cryptosporidiosis with *Cryptosporidium* oocysts in faecal samples of AIDS and other immunosuppressive illness patients, as well as in child diarrhoeal diseases, have been reported in developing countries (Guerrant *et al.* 1990; Guerrant 1997; Sorvillo *et al.* 1998). Epidemic cryptosporidiosis in developed countries probably results from reduced levels of low-dose exposure and an absence of protective immunity to the parasite (Newman *et al.* 1994). On the other hand, cryptosporidiosis in highly endemic areas may occur as a subclinical disease. Even subclinical infections induce serological responses and a number of studies have detected *Cryptosporidium* IgG, IgM and IgA antibodies in different populations (Ungar *et al.* 1986; Casemore 1987; Zu *et al.* 1994). Although individuals from all ages can be infected, clinical manifestations are more frequent in

children and higher seroprevalence in the adult population (Ungar *et al.* 1986; O'Donoghue 1995; Ramirez *et al.* 2004). However, the similar and high prevalence rates of *Cryptosporidium* antibodies found among different age groups and genders in this work, and the absence of clinical cases of cryptosporidiosis among the children's relatives, may reflect the persistent presence of the parasite in the environment and asymptomatic infection occurring repetitively in new susceptible individuals. Although unstable, this herd immunity may protect the population against the occurrence of epidemics. To the best of our knowledge, no massive human cryptosporidiosis outbreaks have been reported in Brazil, except for one recent study involving children attending a day care in the city of São Paulo (Gonçalves *et al.* 2006). Therefore, the very few cryptosporidiosis outbreaks in Brazil may be related to the high levels of serological responses associated with continuous exposure to poor quality of drinking water, as proposed by some authors (Frost *et al.* 2005).

Modes of transmission other than those related to drinking water may be important in some areas, such as the presence of domestic animals and local sanitary conditions (Newman *et al.* 1993). In our study, *Cryptosporidium* antibodies in individuals living in an area without environmental sanitation intervention differed from those individuals living in areas with some environmental sanitation intervention, suggesting that, contamination of surrounding environment may play an important role in *Cryptosporidium* transmission. This fact was expected as this parasite is remarkably resistant to adverse environmental conditions and, due to lack of space and leisure areas in their residences, the studied population (mostly youngsters) frequently circulate in the streets. The children could potentially become infected by playing in an environment contaminated by domestic animals and human faeces. In conclusion, the data presented in this work suggest that *Cryptosporidium* transmission in a periurban area has the environmental conditions, including water, as an important transmission source. However, the household and surrounding environmental contamination may also provide this parasite a permanent circulation among this population, each individual being infected several times.

As cryptosporidiosis outbreaks are not common in Brazil, the major concern is the threat to the health of the HIV-seropositive patients and to malnourished under 5-year-old from low income populations, who can develop severe acute or persistent/chronic diarrhoea illnesses. Because of the lack of an efficient drug therapy, the control of cryptosporidiosis in these patients relies only on the host immune response and on measures to prevent infection, which should emphasize the importance of improving drinking water treatment practices and the establishment

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of surveillance programs for detection of *Cryptosporidium* in the human water supply. Moreover, occurrence of undetected epidemics in Brazil could not be excluded, as the public health system of developing countries might not be well skilled to promptly detect such epidemics.

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Etude sérologique de la transmission du *Cryptosporidium* dans une zone périurbaine d'une ville du nord-est du Brésil

OBJECTIFS Etudier la prévalence de l'infection par le *Cryptosporidium* en mesurant les taux d'anticorps IgG anti cryptosporidium chez les personnes habitant trois quartiers d'une zone périurbaine de Salvador, dans le nord-est du Brésil et, investiguer les effets des mesures d'hygiène environnementale, des habitudes hygiéniques et de l'approvisionnement, du stockage et de l'usage de l'eau de ménage sur la fréquence de ces anticorps sériques dans la population étudiée.

MÉTHODES La transmission inter ménage du *Cryptosporidium* a été étudiée en comparant la fréquence des anticorps IgG anti cryptosporidium chez les personnes habitant des régions avec ou sans mesures d'hygiène environnementales et la transmission intra ménage en comparant la présence de ces anticorps dans des familles avec ou sans cas de diarrhée liées à la présence d'oocystes de *Cryptosporidium* dans les selles. Les enfants ou membres de famille avec des épisodes de diarrhée ont été évalués parasitologiquement pour l'infection au *Cryptosporidium* en examinant des spécimens de selles par la méthode modifiée de Ritchie basée sur la concentration au formol/éther et la méthode de coloration acido-résistante. Tous les groupes ont été sérologiquement évalués pour l'exposition au parasite par une analyse ELISA.

RÉSULTATS Une différence statistiquement significative a été détectée pour la prévalence de l'infection au *Cryptosporidium* entre la zone 1 qui n'avait aucune mesure d'hygiène environnementale et la zone 3 où ses des mesures d'hygiène environnementale avaient été améliorées ($P = 0,044$). La plupart des habitudes hygiéniques étudiées n'ont pas corrélés avec la présence des anticorps sériques anti cryptosporidium dans la population étudiée.

Cependant, des associations positives ont été trouvées autant entre l'approvisionnements pauvre en eau des ménages (OD = 0,17; IC90%: 0,09–0,32; $P = 0,0001$) et la boisson d'eau non bouillie/non filtrée (OD = 0,40; IC90% : 0,24–0,67; $P = 0,0002$) avec des taux élevés d'anticorps sériques anti cryptosporidium.

CONCLUSION Ces données suggèrent que malgré un approvisionnement non approprié en eau de ménage, le stockage et l'usage jouent un rôle important dans la transmission du *Cryptosporidium* dans les secteurs périurbains des villes de pays en voie de développement comme au Salvador, au Brésil. Des conditions environnementales inadéquates peuvent également contribuer à la propagation du parasite.

mots clés *Cryptosporidium*, séoprévalence, transmission, mesures d'hygiène environnementale, approvisionnement en eau

M. C. A. Teixeira *et al.* **Serological study of *Cryptosporidium* transmission****Estudio serológico de transmisión de *Cryptosporidium* en un área peri-urbana de una ciudad al noroeste de Brasil**

OBJETIVOS Estudiar la prevalencia de la infección por *Cryptosporidium*, midiendo los niveles de anticuerpos IgG anti-*Cryptosporidium* entre personas que viven en tres vecindarios de un área peri-urbana de Salvador, al noreste de Brasil; e investigar los efectos que sobre la frecuencia de estos anticuerpos en sueros de la población de estudio tienen las medidas de saneamiento ambiental, los hábitos higiénicos y el manejo y almacenamiento del agua en los hogares.

MÉTODOS La transmisión entre hogares de *Cryptosporidium* se estudió comparando la frecuencia de los anticuerpos IgG anti-*Cryptosporidium* entre personas que habitan estas áreas con o sin diferentes medidas de saneamiento ambiental y transmisión intra-hogares, comparando la presencia de estos anticuerpos en familias con o sin casos de diarrea, asociados a la presencia de oocitos de *Cryptosporidium* en heces. Los niños o miembros de la familia con episodios de diarrea eran evaluados parasitológicamente para infección por *Cryptosporidium* mediante un examen de heces con los métodos de concentración de formol-éter (Ritchie) y el de tinción ácida rápida. Todos los grupos se evaluaron serológicamente para exposición al parásito, utilizando una prueba de ELISA.

RESULTADOS Se detectó una diferencia estadísticamente significativa en la prevalencia de infección por *Cryptosporidium* entre el área 1, que no tenía medidas de saneamiento ambiental, y el área 3 que contaba con mejoras en las medidas de saneamiento ambiental ($P = 0.044$). La mayoría de los hábitos higiénicos estudiados no se correlacionaban con la presencia de anticuerpos anti-*Cryptosporidium* en sueros de la población de estudio. Sin embargo, se encontraron asociaciones positivas entre el tener un contenedor de agua en malas condiciones en el hogar (OD = 0.17; 90% IC = 0.09–0.32; $P = 0.0001$) y el beber agua sin hervir / sin filtrar (OD = 0.40; 90% IC = 0.24–0.67; $P = 0.0002$) con altos niveles de anti-*Cryptosporidium* en suero.

CONCLUSIONES Estos datos sugieren que aunque un almacenamiento y manejo inadecuado del agua en el hogar juega un papel importante en la transmisión de *Cryptosporidium* en las áreas peri-urbanas de ciudades de países en vías de desarrollo, como Salvador, Brasil, las condiciones ambientales inadecuadas también pueden contribuir a la dispersión del parásito.

palabras clave *Cryptosporidium*, seroprevalencia, transmisión, medidas de saneamiento ambiental, suministro de agua