Prevalence of Intestinal Encephalitozoonosis in Mexico

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Infections with *Encephalitozoon* (phylum Microspora) protozoa in immunodeficient patients have emerged worldwide. The prevalence of infections due to *Encephalitozoon* species in nonclinical populations remains unclear. We conducted a cross-sectional survey of two rural highland villages in Mexico by using monoclonal antibody 3B6 to *Encephalitozoon* in immunofluorescence to assess the prevalence of *Encephalitozoon* spores in human stools. We found that 20 (7.84%) of the 255 subjects were positive and that 15 (21.4%) of the 70 households had at least one member who was positive. These results suggest that *Encephalitozoon* species, most likely *Encephalitozoon intestinalis,* may be commonly present in community settings.

The intestinal microsporidial parasite *Encephalitozoon* (*Septata*) *intestinalis* is an important cause of diarrheal illness in patients with AIDS [1–5]. Little is known about its epidemiology, in part because of limited diagnostic methods for routine clinical testing and fieldwork. We recently produced a genus-specific monoclonal antibody (mAb) to *Encephalitozoon* species, named 3B6, that strongly recognizes fresh or fixed spores [6]. We conducted a cross-sectional survey of two rural highland villages in Mexico by using mAb 3B6 in immunofluorescence and found the prevalence of intestinal microsporidial *Encephalitozoon* spores in children and adults.

Materials and Methods

This cross-sectional study was conducted in the summer and fall of 1996 in two rural agricultural communities (San Jose el Rincón and Libertad Tecola) located in central Mexico, 30 km from Puebla City, Puebla. This highland region, surrounded by low mesas, has a moderate climate with a rainy summer season (average rainfall, 200.5 mm/mo).

The communities were surveyed. All households were visited door-to-door. The households were revisited when members were not present before they were considered nonpartici-

pants. The adult respondent of each household was informed of the study's purpose and signed a written informed consent.

Of 116 households, a total of 70 were recruited. Forty-six households not recruited either were not at home (44) or refused to participate (two). Respondents were interviewed, and information obtained included water source, waste disposal, dwelling characteristics, domestic animals, and personal information (age, sex, and hygiene habits). Stool samples were collected from 255 individuals (151 participants from 44 households in San Jose el Rincón; 104 participants from 26 households in Libertad Tecola).

Five milliliters of fecal material was collected from each subject; the samples were fixed in 10% formalin and concentrated by ethyl acetate sedimentation. Concentrates were mounted on polylysine-coated, 10-well diagnostic slides; blocked with bovine serum albumin; and incubated sequentially with mAb 3B6 to *Encephalitozoon* [6] and with fluorescein isothiocyanate–labeled goat antibody to mouse IgG (Kirkegaard and Perry Laboratories, Gaithersburg, MD).

Results were analyzed by using Epi-Info Version 6.0 (Centers for Disease Control and Prevention, Atlanta). Analyses were conducted to determine prevalence. The χ^2 test was used to determine which characteristics were associated with the presence of fecal *Encephalitozoon* spores.

Results

Encephalitozoon spores were identified in 20 (7.84%) of 255 stool samples (table 1). Positive subjects belonged to 15 of the 70 households surveyed. Eight of the 20 positive subjects tested were clustered in three of the 15 households.

There were no significant differences in age between positive and negative individuals ($P > .05$). Of the positive subjects 72.2% were females and 27.8% were males.

Household characteristics (including type of floor, walls, and roofs) were not significantly different between positive and negative households (table 2). Use of water from an indoor faucet was significantly higher in positive households (40%)

Table 1. Percentage of individuals and households with members who had *Encephalitozoon* spores in feces in two rural highland villages in Mexico.

	No. with *Encephalitozoon* spores in stools/total no. (%)	
Village	Individuals	Households
San Jose el Rincón	9/151 (6.0)	7/44 (15.9)
Libertad Tecola	11/104 (10.6)	8/26 (30.8)
Total	20/255 (7.84)	15/70 (21.4)

than in negative households (15.6%; $P < .05$). Unboiled faucet water was used for preparing food or drinking in a greater percentage of positive households than in negative households (table 2; $P < .05$). The source of drinking water, regardless of domestic water treatment, appeared to be a contributing factor to the observed differences between positive and negative households. Forty percent of households with *Encephalitozoon*-positive individuals obtained drinking water from community wells, compared with 25.4% of households with negative members ($P < .05$). In addition, only 20.0% of households with *Encephalitozoon*-positive members obtained their drinking water from a private well, compared with 51.0% of households with *Encephalitozoon*-negative members ($P < .05$).

Discussion

To our knowledge, this is the first study exploring the presence of fecal *Encephalitozoon* spores in whole communities instead of patients in a clinical setting [7–9]. A significant proportion of individuals in this cross-sectional study (7.84%) had *Encephalitozoon* spores in stool samples. This prevalence may reflect intrinsic socioeconomic and hygiene standards that might favor fecal-oral transmission of gastrointestinal pathogens.

Since only one stool sample per participant was collected, any unforeseen variations in fecal excretion of spores within and between days may vary our interpretations. Of all the socioeconomic and hygiene variables studied, only the source and use of water appeared to influence the presence of spores in stools. Individuals from households using faucets or community wells as water sources appeared to be at a greater risk of infection. Households in Libertad Tecola that received piped water had a greater risk of infection than did households in San Jose el Rincón that were using well water. The piped water system of Libertad Tecola is served by untreated water from a spring 5 km away. Water sources in both towns may have been contaminated, but well water may have been less contaminated than piped water. Study of such waters warrants investigation.

It was not the aim of this study to identify people with diarrhea. However, only two of the 255 participants had formed stools. Most people interviewed experienced more than three

evacuations per day. When asked, all respondents claimed not to have had decreased stool consistency in the week before the interview. This high prevalence of unformed stools, commonly seen in developing countries, may indicate comorbidities with

Table 2. Household characteristics for individuals with or without *Encephalitozoon* spores in feces in two rural highland villages in Mexico.

Characteristic	No. (%) of households with characteristic ($n = 70$)	No. (%) of households with characteristic and *Encephalitozoon* spores
Floor construction		
Dirt	10 (14.2)	3 (30.0)
Cement	47 (67.1)	12 (25.5)
Tile	3 (4.3)	0
Other	9 (12.8)	0
Missing	1 (1.4)	0
Wall construction		
Adobe	6 (8.6)	0
Wood	1 (1.4)	1 (100)
Brick	38 (54.3)	10 (26.3)
Cement block	13 (18.6)	3 (23.1)
Other	12 (17.1)	1 (8.3)
Roof construction		
Tin	21 (30.0)	2 (9.5)
Wood	1 (1.4)	0
Cardboard	1 (1.4)	0
Cement block	14 (20.0)	3 (21.4)
Brick	5 (7.1)	0
Other	28 (40.0)	10 (35.7)
Kitchen stove		
Wood	36 (51.4)	10 (27.8)
Gas	18 (25.7)	3 (16.7)
Electric	1 (1.4)	0
Other	15 (21.4)	2 (13.3)
Water source*		
River	2 (2.8)	0
Community well	20 (28.6)	4 (20.0)
Private well	30 (42.8)	3 (10.0)
Faucet	17 (24.3)	7 (41.2)
Bottled	1 (1.4)	1 (100)
Waste disposal		
None	51 (72.8)	12 (23.5)
Latrine	9 (12.8)	2 (22.2)
Indoor toilet	8 (11.4)	1 (12.5)
Other	2 (2.8)	1 (50.0)
Electric lights	69 (98.6)	15 (21.7)
Refrigerator	6 (8.6)	0
Water use		
Preparation of food*		
Unboiled well water	33 (47.1)	4 (12.1)
Unboiled faucet water	19 (27.1)	7 (36.8)
Boiled water	9 (12.8)	2 (22.2)
Other	9 (12.8)	2 (22.2)
Drinking*		
Unboiled well water	26 (37.1)	3 (11.5)
Unboiled faucet water	15 (21.4)	7 (46.7)
Boiled water	14 (20.0)	2 (14.3)
Other	15 (21.4)	3 (20.0)

* $P < .05$ for prevalence of fecal *Encephalitozoon* spores in households with different types of water source and use (χ^2 test).

other intestinal pathogens. Our population, demographics, location, and sampling differed significantly from most reports of microsporidial infections. Nonetheless, we demonstrated that *Encephalitozoon* spores were present in a significant proportion of stool samples from subjects whose risks were not primarily immunodeficiency [2–4, 7–9] but who might have had, among other factors, concomitant intestinal pathogens, poor hygiene, and/or contaminated water.

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