
The Seroprevalence of Cysticercosis, Malaria, and *Trypanosoma cruzi* Among North Carolina Migrant Farmworkers

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Synopsis.....

A seroprevalence study of cysticercosis, Trypanosoma cruzi, and plasmodia species and screening

THE MAJORITY of the more than 4 million migrant farmworkers in the United States (1) were born in Latin America, most in areas endemic for cysticercosis, malaria, and Chagas' disease (*Trypanosoma cruzi* infection). Aside from case studies, there are little or no data available about the prevalence of these infections in this population.

Cysticercosis is the most common parasitic disease of the central nervous system in the United States (2). In Mexico, the home country for most farmworkers, mortality from cysticercosis exceeds that of diabetes or hepatitis (3). Several recent cases of symptomatic, autochthonous cysticercosis have been reported in the United States (4). Transmission of *T. cruzi* occurs in two-thirds of Mexico (5), and a number of autochthonous cases of Chagas' disease have occurred in the United States

for active malaria was conducted among a randomly selected group of 138 Hispanic and Haitian migrant farmworkers. A random sample of labor camps in eastern North Carolina was selected. Blood samples were tested by Indirect Fluorescent Antibody techniques for plasmodial antibody and by enzyme-linked immunosorbent assay (ELISA) for cysticerci and T. cruzi antibodies. Questionnaires collected demographic data and medical history of the workers and family. Blood films stained with Leukostat stain were examined for plasmodia species. The seroprevalence of cysticercosis was 10 percent, T. cruzi 2 percent, and plasmodia species 4.4 percent. One case of active malaria (Plasmodium vivax) was demonstrated.

The clinical significance of seropositivity was not determined, but these results suggest that a small but significant number of farmworkers are infected with cysticercosis, T. cruzi, and malaria. Migrant health clinicians should be aware of the possible presence of these infections. Greater observance and enforcement of sanitation regulations in farmwork is needed to prevent transmission of cysticercosis.

(6-8). Malaria is endemic throughout southern Mexico, Haiti, and Central America (9-14). The five outbreaks of malaria in the United States since 1970 have all occurred among farmworkers (15-18). A seroprevalence study of cysticercosis, *T. cruzi*, and malaria was conducted in 1988 among a randomly selected group of Hispanic and Haitian migrant farmworkers in North Carolina.

Methods

Sample design and subject selection. A 5 percent random sample of migrant camps in eastern North Carolina was selected (SAS Ranuni function). All foreign-born migrants and their family members within selected camps were eligible subjects. A small monetary incentive was offered to minimize

selection bias. Informed consent was obtained and verbal questionnaires were administered in the subject's preferred language (English, Spanish, or Haitian Creole).

Blood collection and examination. Blood was obtained by venipuncture. A thick and thin film were prepared from each blood sample, stained (Leukostat stain), and examined microscopically for plasmodia.

Cysticercosis ELISA. Enzyme-linked immunosorbent assay (ELISA) techniques for cysticercosis antibody testing were performed following previously published protocols (19). Negative controls consisted of dilution buffer only and serums from nine persons born in the United States without history of travel to endemic areas. Several of the serums identified as positive for cysticercosis antibody by the method mentioned previously were tested at the Centers for Disease Control and Prevention (CDC) using an enzyme-linked immunoelectrotransfer blot assay (20).

Malaria. Indirect immunofluorescent antibody (IFA) testing was employed (21,22). Serums were tested against *Plasmodium falciparum*, *Plasmodium vivax*, and *Plasmodium brasilianum* (the antigenic equivalent of *P. malariae*) antigen (23). Three negative controls consisted of two known negative serums and a saline control. Significant reactions at a titer greater than 1:64 (by fluorescence microscopy) were tested again up to a 1:4096 dilution as described earlier. Presumptive positive serums, negative serums, and controls were read blind an additional time by a different researcher. A positive serum sample was defined as one in which significant fluorescence was detected at a titer greater than 1:64 (24).

***T. cruzi* ELISA.** Antigen consisted of glutaraldehyde-fixed LIT culture forms of *Trypanosoma cruzi* (1.5 X 10⁷ per milliliter). Blocking solution consisted of 2.5 percent nonfat dry milk (NFDM) in phosphate buffered saline (PBS) (1 hour, 37° centigrade [C]). Serums were diluted 1:10 in NFDM per PBS, followed by 4x serial dilution in NFDM per PBS (1 hour, 37° C.). Labelling antiserum consisted of goat anti-human gamma-globulin (polyvalent) incubated (diluted 1:1,000 in NFDM per PBS) for 1 hour at 37° C. with 0-phenylenediamine dihydrochloride as substrate. Titer endpoint was defined as the highest dilution of test serum for which the optical density exceeded

the mean control optical density plus two standard deviations and also exceeded 0.05. Four negative controls of normal serums were employed.

Vector study. Mosquito larvae were collected from ponds within anopheline flight range of the camp where the worker with the active case of malaria resided. Three incandescent, carbon dioxide baited, downdraft type traps were used, as well as live baiting with human volunteers. Mosquitoes were identified, dissected, examined for parity and for the presence of oocysts and sporozoites.

Intestinal parasites. As part of a concurrent study of intestinal parasites among farmworkers, most subjects provided stool samples which were screened for intestinal parasites using standard methods (25).

Results

Serums from 138 farmworkers and family members (63 percent Mexican-born, 21 percent Haiti-born, 12 percent Central America-born, 4.5 percent U.S.-born) from 17 migrant camps in four counties in eastern North Carolina were tested for presence of antibody to cysticerci, *P. falciparum*, *P. vivax*, *P. brasilianum*, and 99 for *T. cruzi* (Haitian-born subjects were not tested for *T. cruzi*). Nonresponse was approximately 40 percent of all potential subjects.

Cysticercosis. Seroprevalence was 10 percent of the total group (14) and 12 percent (10) among the Mexican-born (see table). Seropositive serums were confirmed by replicate ELISA testing. The single U.S.-born farmworker who was seropositive for cysticercosis, an Hispanic male, reported no trips outside the United States. Persons from Guanajuato comprised 13 percent of Mexican-born group but 30 percent of the seropositive Mexican-born. Seropositive persons averaged twice the number of reported exits from the United States to their country of origin compared with the seronegative ($P = .012$, *T*-test).

Sixty-six percent of the group submitted stool samples which were examined for intestinal parasites. No eggs of *Taenia* species were found. The prevalence of intestinal parasites was not significantly higher among seropositives (60 percent) than seronegatives (54 percent) ($P = 1.00$, Fisher's exact test).

Seropositivity was not associated with previous history of parasitic infection ($P = .329$, Fisher's

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exact test) or with reported infection of family members ($P = .410$, Fisher's exact test).

Ten of the 14 who were serum-positive by ELISA were retested by immunoblot at CDC (21). Four of these serums were positive by this test.

Malaria. Of the subjects, 28 percent reported that malaria was common in their home region: 44 percent of Central Americans, 29 percent of Mexicans, and 6 percent of Haitians. Previous infection with malaria and family members' or acquaintances' infections were also frequently reported (8–38 percent). The seroprevalence of plasmodial infection in this sample was 4.4 percent—five positive for *P. falciparum* and one for *P. vivax* (see table). All persons who were seropositive for *P. falciparum* were Haitian-born; the seroprevalence among Haitians was 17 percent. The person seropositive for *P. vivax* had an active infection, with a heavy patent infection (class 4, World Health Organization scale). Upon enrollment he was acutely ill, and three paroxysms had occurred prior to his diagnosis and treatment upon enrollment in the study. He had previously received incorrect treatment in North Carolina (chloroquine was prescribed but not primaquine).

Vector study. Large numbers of anopheline larvae were identified in one pond within anopheline flight range (less than one-fourth mile) of the labor camp where the person with the active case of malaria resided. Of the adult mosquitoes obtained from the overnight traps, 40 percent were *Anopheles*, primarily *A. quadrimaculatus* and a few *A. pseudopunctipennis*. All females were nulliparous upon dissection, and no sporozoites or oocysts were found. Collections of biting insects were performed on subsequent nights with similar results.

T. cruzi. Serums from 99 subjects were tested for antibody to *T. cruzi* (Haitian subjects were not included, and serums from 10 subjects were exhausted by the previous multiple serologies). Two Mexican-born subjects were seropositive, with titers of 1:40,960 and 1:10,240.

Discussion

Given the prevalence of these infections in foreign-born farmworkers' home regions, both seropositivity and active infections are predictable in a sufficiently large sample. Unfortunately, we could not accomplish serologic testing until several weeks after collection of serums and were then unable, despite weeks of searching, to locate subjects with positive cysticercosis and *T. cruzi* titers.

The small sample size and high nonresponse rate make it difficult to extrapolate these findings to the population of foreign-born farmworkers. Use of a monetary incentive probably reduced selection bias, but it is difficult to predict the direction of possible bias in nonrespondents. However, we found that subjects less familiar with health care—those from more rural and remote areas—were more likely to nonrespond. Thus, underestimation of prevalence may have occurred.

Cysticercosis constitutes the most medically significant infection studied. Considerable effort has been devoted to developing serologic tests (21,26–34), and cross-reactivity has been a major problem (28,35–37). The ELISA test used in this study has been found to be both sensitive and specific (19). False-positives are most common with antigenically similar helminths (*Echinococcus granulosis* and *Echinococcus multilocularis*), largely absent from Mexico and Central America (28,38). The presence of seropositive subjects was confirmed by immunoblot testing at the CDC, although the discrepancies between the results of the two testing procedures raises the possibility that some false positives may have been obtained by ELISA testing. The immunoblot studies cannot, however, be used to adjust the seropositivity rate, since only a fraction of all serums were tested by immunoblot. Even if only the immunoblot confirmed serums were counted as true positives, a seroprevalence of 3 percent would, if extrapolated to the population of approximately 40,000 foreign-born farmworkers in North Carolina, result in an estimate of 1,200 seropositive persons in this one State.

The cysticercosis seroprevalence of 10 percent reported in this study is consistent with studies in

Seroprevalence of cysticercosis and plasmodia species among North Carolina migrant farmworkers

Infective agent	Birthplace									
	Total sample (N = 138)		Haiti (N = 29)		Mexico (N = 87)		Central America (N = 16)		USA (N = 6)	
	Number	Percent	Number	Percent	Number	Percent	Number	Percent	Number	Percent
Cysticerci.....	14	10	2	7	10	12	1	6	1	17
Plasmodia species.....	6	4.4	5	17	1	1	0	0	0	0
<i>P. vivax</i>	1	0.7	0	0	1	1	0	0	0	0
<i>P. falciparum</i>	5	3.6	5	17	0	0	0	0	0	0

the home regions of most subjects (3). However, the seroprevalence of 7 percent among Haitian farmworkers is surprising, since cysticercosis in Haiti is reportedly of low and sporadic incidence (39). No epidemiologic studies of cysticercosis in Haiti have been conducted.

The demonstration of a seropositive case (immunoblot confirmed) among the U.S.-born suggests the possibility of endemic cysticercosis transmission among farmworkers. The number of autochthonous cases reported in the United States (2,40-48) and the transmission of parasites in migrant farmwork (25) underscore the possibility of autochthonous transmission. The conditions permitting the transmission of *Taenia* infection are the same as those which would allow transmission of many other types of intestinal parasites. Several studies have shown that parasite transmission occurs among migrants, and that basic hygienic and sanitation measures such as handwashing water in the fields and field sanitation are not regularly available (25,49-50).

Recently, there have been several cases of autochthonous cysticercosis reported in eastern States (4). One occurred in North Carolina and involved a family member of a family-owned farm whose farmworker employees were the probable source of infection. The transmission of this potentially fatal infection constitutes a serious public health concern and adds further weight to the already substantial body of data demonstrating the need for improved sanitation and hygiene in the workplace and living quarters of migrant farmworkers.

The seroprevalence of *T. cruzi* infection conforms fairly well with Kirchoff's finding of a 4.9 percent seroprevalence among a clinic-based sample of Central Americans (51). The reliability of this figure as a population estimate is limited by the small sample size and low number of positives. However, even if prevalence is overestimated by twofold or threefold, several hundred seropositive Hispanic farmworkers may work in North Carolina. Thus, the possibility of pathology is real.

Transmission of Chagas' disease by blood transfusion, common in Latin America (52-54), has recently become a concern in the United States also (55).

Major surveillance and control efforts have been undertaken for certain groups of immigrants at high risk for malaria, such as Indochinese refugees and Punjabi Sikhs (16,56). However, because of the logistical difficulties involved in surveillance, minimal resources for migrant health, and the presence of numerous, far more serious health problems among this population, surveillance among migrant farmworkers has been nonexistent. Given the prevalence of malaria in regions from which farmworkers originate, it is not surprising that patent parasitemia and seropositivity were demonstrated in this study. Approximately 80 percent of Haiti's population reside in areas of active transmission, and malaria incidence increased in Haiti during the 1980s (57). Thus, the seroprevalence of 17 percent found among Haitians is not surprising. Haitians are the only group in the migrant population from an area of suspected resistance (58,59). However, unlike the other infections studied, the extended persistence of plasmodial seropositivity means that seroprevalence alone would greatly overestimate the occurrence of active infection or risk of transmission.

Based upon one active case of malaria in this small sample, it is obviously difficult to estimate the prevalence in the population. It seems unlikely that the only case imported into North Carolina surfaced in a random sample constituting a small fraction of the population. This subject's experience indicates that treatment for malaria received by migrants can be both delayed and incorrect, increasing the possibility of transmission. We found large numbers of suitable vectors in the area, indicating that transmission was entirely possible.

Comparison with the farmworkers' outbreak in San Diego County, CA, is instructive (17). This outbreak was detected only because a nonmigrant became infected. That 26 migrants had patent

parasitemias, undetected until a single nonmigrant became infected, is strong evidence that transmission can continue undetected.

The results of this study have significance on both a clinical and public health level. Some conditions of farmwork, such as the lack of field sanitation and inadequate hygienic facilities, increase the risk for transmission of cysticercosis. The unfamiliarity of U.S. trained physicians with these infections, and farmworkers' isolation, transience, and low access to health care, make nondetection or misdiagnosis possible. Physicians serving migrant populations need to be aware of the seroprevalence of cysticercosis, *T. cruzi*, and malaria demonstrated in this population.

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