ABSTRACT: We tested sera from 176 homeless people in Houston for antibodies against typhus group rickettsiae (TGR). Sera from 19 homeless people were reactive to TGR antigens by ELISA and IFA. Two people had antibodies against Rickettsia prowazekii (epidemic typhus) and the remaining 17 had antibodies against Rickettsia typhi (murine typhus). Journal of Vector Ecology 33 (1): 206-208. 2008.

Keyword Index: Rickettsia, homeless, typhus, lice, flea.

INTRODUCTION

Houston is the fourth largest city in the United States and has a homeless population estimated at 10,000 persons (Health, Hope and Dignity Program Narrative 2007). The urban homeless can be exposed to a wide variety of ectoparasitic arthropods such as fleas and lice (Brouqui et al. 2005, Brouqui and Raoult 2006). Rickettsial diseases caused by typhus group rickettsiae (TGR) are transmitted by these ectoparasites and are endemic to Texas (e.g., Taylor et al. 1986, Azad et al. 1997, Azad and Beard 1998, Fergie et al. 2000). The urban homeless in some cities, such as Marseilles, France, have shown evidence of exposure to TGR (Brouqui et al. 2005). We wanted to determine if the homeless population in Houston, TX, had exposures to TGR and to differentiate cases of murine typhus (Rickettsia typhi) from epidemic typhus (Rickettsia prowazekii) in this population.

MATERIALS AND METHODS

We used sera previously collected for an arbovirus study in Houston (Meyer et al. 2007). We conducted a cross-sectional serological survey using convenience sampling from shelters, soup kitchens, and other outreach organizations serving homeless adults. From March to May, 2004, a total of 426 participants were enrolled from 13 sites; 419 interviews were completed from those enrolled. We tested a random sampling of sera from a total of 176 participants for the presence of antibodies to TGR.

Serum was tested for antibodies reactive with TGR using an immunoglobulin G (IgG) enzyme linked immunosorbent assay (ELISA) and confirmed using an indirect fluorescent assay (IFA). Sera that were reactive with both R. typhi and R. prowazekii by IFA were further differentiated by cross-adsortion. For IgG ELISA, gradient-purified R. typhi was disrupted by pressure cell (Halle and Dasch 1980). Low-binding ELISA microplates (MP Biomedicals, Inc., Irvine, CA) were coated with 1.5 ng of rickettsial protein per well and blocked with 3% bovine serum albumin (BSA) (Sigma, St. Louis, MO) in phosphate buffered saline (PBS). Sera were diluted 1:100 in ELISA buffer (PBS with 1% BSA, 0.01% Tween-20, and 0.02% thimerosol), and 50 µl were applied to duplicate wells. After incubation at 37°C for 90 min, plates were washed three times and 10 ng of horseradish-peroxidase conjugated rabbit anti-human IgG (KPL, Gaithersburg, MD) in ELISA buffer was added to each well. After incubation at 37°C for 60 min, plates were washed four times. One-component ABTS substrate (KPL) was applied and incubated for 15 min; the reaction was stopped with 1% SDS. The optical density at 405 nm was recorded, and sera-producing optical densities greater than 0.60 were considered to be reactive by ELISA. The cut-off value for optical density was based on unpublished analyses of over 100 negative sera.

All sera determined to be reactive by ELISA were verified by IFA. We used ELISA for the initial screening because the technique allows for high throughput screening. IFA was performed as previously described for A. phagocytophilum (Comer et al. 1999a), but with R. typhi (Wilmington strain) grown in VERO cells or R. prowazekii (Breinl strain) grown in egg yolk sac as the antigen. Positive, negative, and diluent controls were used in each test. Sera were screened at a dilution of 1:64. Sera deemed positive at 1:64 were retested at serial dilutions from 1:64 to 1:256, and sera reactive at 1:256 were determined to be positive. Sera that reacted with both R. typhi and R. prowazekii antigens...
were further analyzed using cross-adsorption, as previously described (Comer et al. 1999b).

RESULTS

Sera from 19 (10.7%) of the 176 homeless people tested were determined to be reactive with TGR antigens by ELISA and IFA. Of the 19 positive sera, 17 (89%) were determined to be reactive with \( R. typhi \), and two (11%) were determined to be reactive with \( R. prowazekii \).

Of the two positive participants for \( R. prowazekii \), one was a 34-year-old African-American, non-Hispanic male with a history of living three years in Mexico. He described himself as living in temporary emergency shelters two to three times per week and had been homeless for over 16 years. The other \( R. prowazekii \) positive individual was a 54-year-old Caucasian, non-Hispanic female with no history of living outside the United States. She was living in a transitional living facility and had been homeless for four to five months.

All of the 17 homeless individuals who were positive for \( R. typhi \) were born in the United States, with four (24%) having a history of living outside the United States (Germany, West Africa, and Mexico). Eleven (65%) of the 17 positives were male; ten were African-American, six were Caucasian, and one was Hispanic. Median age was 46 years, with a range of 27 to 69 years. Five (29%) of the 17 described themselves as mostly living on the streets, four (24%) lived mostly in temporary emergency shelters, three (18%) lived mostly with family and friends, and five (29%) listed other housing arrangements. Five (29%) reported being homeless for greater than one year and six (35%) stated spending all day and all night outdoors.

DISCUSSION

Our data indicate that a subset of the homeless population in Houston was exposed to \( R. typhi \) (murine typhus) and \( R. prowazekii \) (epidemic typhus). Murine typhus is rarely fatal in humans, and cases are reported from Texas yearly (Traub et al. 1978, Taylor et al. 1986). Rat fleas (\( Xenopsylla cheopis \)) are considered the primary vectors of \( R. typhi \) to humans (Traub et al. 1978). Rats are urban animals and the urban homeless could have an increased risk of exposure to both rats and fleas. Since murine typhus has a wide geographic distribution, we cannot determine where the sero-positive people were exposed. In the U.S., murine typhus tends to be restricted to the southern portions of the country from California to Texas (Wiggers and Stewart 2002); however, antibodies against \( R. typhi \) were recently reported in rats from northern cities such as Baltimore (Traub et al. 1978, Reeves et al. 2006). Murine typhus is not transmitted from human to human. Similar seological surveys in Texas have reported positive sera for exposure to \( R. typhi \) from approximately 9% to 16% of selected non-homeless populations (Wiggers and Stewart 2002, Purcell et al. 2007).

Brouqui et al. considered a positive titer of 1:64 to be indicative of infection for murine typhus (Brouqui et al. 2005). We used 1:256 as a more conservative cut-off. With these stricter cut-off criteria, we acknowledge that some sero-positive people could be reported as negative. However, \( R. felis \), a spotted fever group \( Rickettsia \), also causes rickettiosis in Texas, and Zavala-Velazquez et al. (1999) reported weak cross-reactivity at 1:64 between antibodies against \( R. felis \) and \( R. typhi \). We considered a titer of 1:256 to be the minimum to rule out cross-reactivity with \( R. felis \). If we had used a 1:64 dilution, an additional eight sera would have been positive for TGR. We did not use \( R. felis \) as an antigen, because of the limitations in culturing high densities of this \( Rickettsia \) for antigen preparations.

Two of the homeless were sero-positive against \( R. prowazekii \), the agent of louse-borne epidemic typhus, which was very concerning. We confirmed this result by cross-adsorption using \( R. typhi \) and \( R. prowazekii \). Acute, untreated cases of epidemic typhus are often fatal, and chronic infections are serious (known as Brill-Zinsser disease). With only having access to a single serum sample per individual, we were unable to determine their disease status. The presence of sero-positive individuals should be a warning that epidemics are possible. An epidemic could be initiated if louse-infested infected and uninfected people are crowded in unsanitary living conditions, such as those that can exist in some homeless camps and shelters.

Typhus group rickettsiae continue to be threats to public health in Texas. Louse-borne diseases such as epidemic typhus, trench fever, and louse-borne relapsing fever have emerged in the homeless populations of some European cities (Brouqui et al. 2005) and public health officials should be aware of their presence or potential emergence in the United States.

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