

# Increased Risk of Tuberculosis With Human T-Lymphotropic Virus-1 Infection

## A Case-Control Study

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**Objectives:** To investigate whether infection with human T-lymphotropic virus type 1 (HTLV-1) increases the risk of tuberculosis.

**Design:** A case-control study.

**Setting:** City of Salvador, Brazil.

**Participants:** A total of 375 patients with tuberculosis (cases) and 378 individuals without tuberculosis (controls), matched by age and sex.

**Main Outcome Measure:** Tuberculosis of lung or lymph node.

**Main Exposure:** Human HTLV-1 infection.

**Results:** The prevalence of HTLV-1 infection was 4.27% (16/375) in patients with tuberculosis and 1.32% (5/378) in controls, resulting in a crude odds ratio of 3.31 (95% CI, 1.20–9.13) and an adjusted odds ratio of 3.01 (95% CI, 1.06–8.58).

**Conclusion:** HTLV-1 infection is associated with a first diagnosis of tuberculosis. This may have implications for tuberculosis control in places with high prevalence of HTLV-1.

**Key Words:** human T-lymphotropic virus type 1, tuberculosis, case-control, Brazil

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The human T-lymphotropic virus type 1 (HTLV-1) was the first human retrovirus identified. It has been clearly implicated in the pathogenesis of adult T-cell leukemia/lymphoma and myelopathy/tropical spastic paraparesis.<sup>1–4</sup> Other infec-

tious, inflammatory, or immune-related diseases (uveitis, infective dermatitis, polymyositis, arthropathy, Sjögren syndrome, interstitial lymphocytic pneumonia) are suspected to be associated with infection with HTLV-1.<sup>5,6</sup> A higher prevalence of HTLV-1 infections has been described in patients with tuberculosis, as well as an increased mortality rate in HTLV-1- and tuberculosis-coinfected patients.<sup>7,8</sup> However, studies addressing this question have been small and failed to provide conclusive evidence of an association.<sup>9,10</sup> The case-control study reported here investigated whether infection with HTLV-1 is associated with increased risk of tuberculosis. The study was conducted in Salvador, a city known to have a relatively high prevalence of HTLV-1 infection (1.76% in the overall population)<sup>11</sup> and a yearly incidence of tuberculosis varying around 120 cases per 100,000 inhabitants.<sup>12</sup>

## PARTICIPANTS AND METHODS

### Study Site and Population

Salvador is the capital city of the state of Bahia, in northeastern Brazil, and is characterized by marked socioeconomic differences. The majority of the estimated population of approximately 2.5 million of inhabitants is of African or mixed African/Portuguese descent. Five of the 12 health districts in Salvador were selected to be included in the study because they had a high incidence of tuberculosis. The estimated population in these 5 districts was 1,230,000 inhabitants, just under half of the total population of Salvador.

The study was approved by the institutional review board of the Hospital Santa Isabel and case and control subjects gave written informed consent before entering the study.

### Cases

Case subjects were recruited from those diagnosed by the Tuberculosis Control Program. The inclusion criteria were a first diagnosis of tuberculosis according to the World Health Organization criteria,<sup>13</sup> confirmed by a senior pulmonologist; duration of treatment not exceeding 6 months prior to recruitment; and age 15 years or older. Exclusion criteria were having had a previous diagnosis of tuberculosis; pregnancy; and presence of HIV infection or any other condition leading to immunosuppression, including suspected or confirmed cancer and collagenosis and treatment with systemic corticosteroids.

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## Controls

Control subjects were selected from the outpatient clinic of the Hospital Santa Isabel, matched to case subjects by sex and age (in 10-year age bands). This hospital is part of the National Public Health Service, and people living in its catchments have similar socioeconomic and demographic characteristics as those living in the areas where the patients with tuberculosis were recruited. The inclusion criteria included absence of tuberculosis; residence in the city of Salvador; and age 15 years or older. The exclusion criteria included pregnancy and presence of HIV infection or other conditions associated with a state of immunosuppression including suspected or confirmed cancer, collagenosis, and treatment with systemic corticosteroids.

## Data Collection

Demographic information (age, sex), socioeconomic status (income and level of education), lifelong number of sexual partners, recreational drug use, and history of blood transfusion were collected by standard questionnaire, which was applied to all individuals participating in the study. Ten milliliters of blood were withdrawn from each individual in ethylene diamine tetra-acetic acid-containing sterile tubes. The plasma was separated through centrifugation and both plasma and blood cells were stored at  $-20^{\circ}\text{C}$ .

## Laboratory Methods

Plasma samples were screened for antibodies to HTLV-1 and 2 (HTLV-I/HTLV-II Ab-Capture ELISA Test System, Ortho Clinical Diagnostic Inc., Raritan, NJ) and HIV-1 and 2 (HIV-1/HIV-2 Ab-Capture ELISA Test System, Ortho Clinical Diagnostic). All samples with positive results for HTLV or HIV in the 1st test were tested again in duplicate. In case of positive results, on the 2nd test the samples were submitted for serologic confirmation. Confirmation of HIV-1 infection was performed by indirect immunofluorescence assay (Biomanguinhos Fiocruz, Rio de Janeiro, Brazil) following the manufacturer's recommendations. For HTLV the confirmation and discrimination between HTLV-1 and HTLV-2 were performed using Western blot analysis (HTLV Blot 2.4; Genelabs, Singapore). Polymerase chain reaction analysis was performed in samples with undetermined HTLV serologic status as described elsewhere.<sup>14</sup>

## Statistical Analysis

The main dependent variable was being a case of tuberculosis and the main exposure of interest was seropositivity for HTLV-1. Univariable analysis was explored using the Pearson  $\chi^2$  test with no continuity correction, with Yates correction, or Fisher exact test; and for ordered or asymmetric, continuous variables, Mann-Whitney  $U$  test. Multivariable analysis was used to investigate the association between HTLV seropositivity and tuberculosis, adjusting for potential confounding variables, using unconditional logistic regression keeping age and sex in the model. This analytical approach is recommended for frequency matching and is equivalent to a conditional logistical regression in which the matching sets are large and include all the cases and all the controls on the same category of the matching variables, in this case of the

same sex and age group. The final model was selected by incorporating variables based on the "likelihood ratio" with  $\alpha = 0.05$ . We present odds ratios (ORs) and 95% CIs. All analysis were done using the statistical software STATA 7.0 (Statacorp., College Station, TX) and SPSS version 11.0 (SPSS, Inc., Chicago, IL).

## RESULTS

A total of 753 individuals were enrolled in this study between September 2001 and October 2002. There were 389 case subjects and 378 control subjects; 14 individuals in the case group were excluded because they were infected with HIV (3 of whom were also infected with HTLV-1). The median age was 34 years (range, 15–84 years) in cases and 35 years (range, 15–84 years) in controls. Cases had lower levels of education and income and more frequent history of using intravenous illicit drugs than controls (Table 1). The majority of cases (59%) were recruited within 3 months of the start of their treatment for tuberculosis, and 20% at the time of diagnosis (data not shown). No individual in the control group was excluded because of infection with HIV.

Sixteen of 375 individuals in the case group were infected with HTLV-1, compared with 5 of 378 individuals in the control group. Giving a crude OR 3.31 (95% CI, 1.20–9.13); this remained substantial and statistically significant (OR 3.01; 95% CI, 1.06–8.58) after controlling for age, sex, education, income, ethnicity, sexual history, and history of blood transfusion and intravenous drug use (Table 2).

## DISCUSSION

We report here a statistically significant increase in the risk of tuberculosis in subjects infected with HTLV-1. Previous studies have investigated this association, but their results have been inconclusive: Matsuzaki et al<sup>15</sup> described an increased reported history of tuberculosis in 2847 subjects infected with HTLV-1 in Japan. A case-control study in Senegal found no significant association between tuberculosis and HTLV-1,<sup>9</sup> but the power of the study was small.<sup>5</sup> A study comparing 154 HTLV-1-infected individuals with 799 uninfected blood donors concerning their history of infectious diseases, including tuberculosis, showed an OR of 3.3 (95% CI, 0.8–14.2).<sup>10</sup>

The prevalence of infection with HTLV-1 reported here (4.27%) is similar to that described in outpatients with tuberculosis from Nigeria (3.6%).<sup>16</sup> The prevalence found in cases of tuberculosis in the present study was lower, when compared with previous surveys in cases of tuberculosis in the same area.<sup>7,8</sup> In both previous papers, hospitalized cases of tuberculosis were studied, as opposed to the study reported here, which included outpatients with a 1st episode of tuberculosis. HTLV-1 has been shown to be associated with a worse course of tuberculosis, with lethality up to 25% in those infected with HTLV-1 compared with 8% in those not infected.<sup>8</sup> This would lead to a higher prevalence in hospitalized cases.

The association we found between HTLV-1 and tuberculosis is biologically plausible, as a consequence of the immunologic deficiencies associated with infection by HTLV-1,<sup>17</sup> which would facilitate multiplication of the *Mycobacterium* and

**TABLE 1.** Sociodemographic and Health Characteristics of Tuberculosis Cases and Controls, Salvador, Bahia, Brazil

Variable	Cases		Controls		P
	n = 375	%	n = 378	%	
Average age, y	36.19		36.71		0.57
Sex					0.31
Male	221	58.9	209	55.3	
Female	154	41.1	169	44.7	
Race					0.001
White	48	12.8	82	21.7	
Nonwhite	327	87.2	296	78.3	
Education/years					<0.001
Illiterate	23	6.1	23	6.1	
Incomplete Jr. high school	193	51.5	127	33.6	
Jr. high school completion	62	16.5	57	15.1	
Incomplete high school	91	24.3	162	42.9	
High school completion or higher	6	1.6	9	2.4	
Income (minimum wage)*					<0.001
<1	81	21.6	36	9.5	
1	160	42.7	152	40.2	
2-3	93	24.8	123	32.5	
4-9	28	7.5	52	13.8	
≥10	7	1.9	15	4.0	
Unknown	6	1.6	—	—	
Blood transfusion					0.006
Yes	16	4.2	36	9.5	
No	358	95.5	338	89.4	
Unknown	1	1.3	4	1.1	
Drug use					<0.001
No	329	87.7	368	97.4	
Intravenous drug	15	4.0	0	0.0	
No intravenous drug	31	8.3	10	2.6	
Sexually active life past or current					<0.001
Yes	232	61.9	279	73.8	
No	142	37.8	96	25.4	
Unknown	1	0.3	3	0.8	
Lifelong number of sexual partners					0.71
1	104	27.7	110	29.1	
2	67	17.9	55	14.6	
3	35	9.3	42	11.1	
4-10	61	16.3	71	18.8	
>10	66	17.6	69	18.3	
Unknown	42	11.2	31	8.2	
Diabetes (referred)	23	6.1	19	5.0	0.50

\*Minimum wage = US \$82.

**TABLE 2.** Risk of Tuberculosis in HTLV-1–Infected Individuals, Salvador, Bahia, Brazil

	OR	95% CI	P Value
HTLV-1 (+)	3.01	1.06–8.58	0.039
Race	1.82	1.21–2.76	0.004
Education (<Jr. high school)	2.07	1.51–2.83	<0.001
Blood transfusion	0.41	0.22–0.78	0.007
Sexual activity	0.54	0.39–0.75	<0.001
Intravenous drug use	3.53	1.68–7.43	0.001

Excluded from the final model: age ( $P = 0.16$ ), sex ( $P = 0.30$ ), income ( $P = 0.06$ ), lifelong number of sexual partners ( $P = 0.30$ ), and diabetes ( $P = 0.24$ ).

CD4CD45RO<sup>+</sup> cell subset.<sup>23</sup> Impairment of the T-cell function is likely to occur: T lymphocytes from HTLV-1–infected individuals vaccinated with bacillus Calmette-Guerin fail to respond to purified protein derivative stimulation (PPD) in vitro,<sup>24</sup> even in the presence of a strong T-helper type 1 response with high levels of interferon- $\gamma$ .<sup>25</sup> This is consistent with the evidence that asymptomatic infection with HTLV-1 is associated with reduced skin response to tuberculin (purified protein derivative).<sup>26-31</sup> This has been interpreted as reflecting reduced cellular immunity, which potentially could lead to a higher rate of active tuberculosis in those with tuberculosis infection.<sup>5</sup> Consistent with this hypothesis is the fact that the prevalence of HTLV-1 is higher in patients with pulmonary disease caused by *Mycobacterium avium*<sup>32</sup>; however, the fact that delayed hypersensitivity skin testing to mumps and *Candida albicans* antigens is the same among HTLV-1 and seronegative individuals<sup>33</sup> suggests that the picture is more complex.

We are aware that the main limitation of the present study is the possibility of incomplete control of confounding. Tuberculosis and HTLV-1 have similar risk factors: poverty, low level of education, and intravenous drug use. However, control for these potential confounding variables hardly changed the OR, suggesting that the association is a causal effect. In conclusion, our results suggest that infection by HTLV-1 is significantly associated with a 1st diagnosis of tuberculosis. This would have implications for public health in areas where the prevalence of HTLV-1 is high. We recommend that HTLV-1–infected individuals be monitored for symptoms of tuberculosis, allowing for an early diagnosis and treatment, and that cases of tuberculosis be screened for HTLV-1 in areas of high prevalence of both diseases. Consideration should be given to the potential benefit of chemoprophylaxis in subjects with HTLV-1 infection and strong reaction to purified protein derivative to prevent development of tuberculosis, similar to the recommendations for HIV coinfection.<sup>34</sup>

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progression to clinical disease.<sup>8,18</sup> There is evidence that HTLV-1 infection could lead to immunologic modulation with significant effects on infection with helminths such as *Strongyloides stercoralis* and *Schistosoma mansoni*.<sup>19,20</sup> It has been established that HTLV-1 induces T-lymphocyte spontaneous proliferation in vitro<sup>21,22</sup> that is due predominantly to a memory

## REFERENCES

- Poiesz BJ, Ruscetti FW, Gazdar AF, et al. Detection and isolation of type C retrovirus particles from fresh and cultured lymphocytes of a patient with cutaneous T-cell lymphoma. *Proc Natl Acad Sci USA*. 1980;77:7415–7419.
- Yoshida M, Miyoshi I, Hinuma Y. Isolation and characterization of retrovirus from cell lines of human adult T-cell leukemia and its implication in the disease. *Proc Natl Acad Sci USA*. 1982;79:2031–2035.
- Gessain A, Barin F, Vernant JC, et al. Antibodies to human T-lymphotropic virus type-I in patients with tropical spastic paraparesis. *Lancet*. 1985;2:407–410.
- Osame M, Usuku K, Izumo S, et al. HTLV-I associated myelopathy, a new clinical entity. *Lancet*. 1986;i:1031–1032.
- Marsh BJ. Infectious complications of human T cell leukemia/lymphoma virus type I infection. *Clin Infect Dis*. 1996;23:138–145.
- Manns A, Hisada M, La Grenade L. Human T-lymphotropic virus type I infection. *Lancet*. 1999;353:1951–1958.
- Moreira ED Jr, Ribeiro TT, Swanson P, et al. Seroepidemiology of human T-cell lymphotropic virus type I/II in northeastern Brazil. *J Acquir Immune Defic Syndr*. 1993;6:959–963.
- Pedral-Sampaio DB, Martins Netto E, Pedrosa C, et al. Co-infection of tuberculosis and HIV/HTLV retroviruses: frequency and prognosis among patients admitted in a Brazilian hospital. *Braz J Infect Dis*. 1997;1:31–35.
- Kaplan JE, Camara T, Hanne A. Low prevalence of human T-lymphotropic virus type I among patients with tuberculosis in Senegal. *J Acquir Immune Defic Syndr*. 1994;7:418–420.
- Murphy EL, Glynn SA, Friley J, et al. Increased prevalence of infectious diseases and other adverse outcomes in human T lymphotropic virus types I- and II-infected blood donors. Retrovirus Epidemiology Donor Study (REDS) Study Group. *J Infect Dis*. 1997;176:1468–1475.
- Dourado I, Alcantara LC, Barreto ML, et al. HTLV-I in the general population of Salvador, Brazil: a city with African ethnic and socio-demographic characteristics. *J Acquir Immune Defic Syndr*. 2003;34:527–531.
- Xavier M. *Tuberculose na Cidade de Salvador: Perfil epidemiológico na década de 1990 e Tendências das Taxas de Incidência e Mortalidade Entre 1981 a 2000* [dissertation]. Salvador, Brasil: Federal University of Bahia; 2002.
- Falzon D. Revised international definitions in tuberculosis control: comments from the Aral Sea Area tuberculosis programme. *Int J Tuberc Lung Dis*. 2001;5:1071–1072.
- Bittencourt AL, Dourado I, Filho PB, et al. Human T-cell lymphotropic virus type I infection among pregnant women in northeastern Brazil. *J Acquir Immune Defic Syndr*. 2001;26:490–494.
- Matsuzaki T, Otose H, Hashimoto K. Diseases among men living in human T-lymphotropic virus type I endemic areas in Japan. *Intern Med*. 1993;32:623–628.
- Olaleye DO, Bernstein L, Sheng Z, et al. Type-specific immune response to human T cell lymphotropic virus (HTLV) type I and type II infections in Nigeria. *Am J Trop Med Hyg*. 1994;50:479–486.
- Goon PK, Bangham CR. Interference with immune function by HTLV-1. *Clin Exp Immunol*. 2004;137:234–236.
- Hollberg P. Mechanisms of T-cell activation by human T-cell lymphotropic virus type I. *Microbiol Mol Biol Rev*. 1999;63:308–333.
- Porto AF, Neva FA, Bittencourt H, et al. HTLV-1 decrease Th2 type of immune response in patients with strongyloidiasis. *Parasite Immunol*. 2001;23:503–507.
- Porto AF, Santos SB, Alcantara L, et al. HTLV-1 modifies the clinical and immunological response to schistosomiasis. *Clin Exp Immunol*. 2004;137:424–429.
- Kramer A, Jacobson JF, Reuben EL, et al. Spontaneous lymphocyte proliferation in symptom free HTLV-1 positive Jamaicans. *Lancet*. 1989;ii:923–924.
- Popovic M, Flomenberg N, Volkman D, et al. Alteration of T-cell functions by infection with HTLV-I or HTLV-II. *Science*. 1984;226:459–462.
- Prince HE, York J, Owen SM, et al. Spontaneous proliferation of memory (CD45RO+) and naïve (CD45RO-) subsets of CD4 cells and CD8 cells in human T lymphotropic virus (HTLV) infection: distinctive patterns for HTLV-I versus HTLV-II. *Clin Exp Immunol*. 1995;102:256–261.
- Suzuki M, Dezzutti CS, Okayama A, et al. Modulation of T-cell response to recall antigens in human T-cell leukemia virus type-1 infected individuals. *Clin Diagn Lab Immunol*. 1999;6:713–717.
- Carvalho EM, Bacellar O, Porto AF, et al. Cytokine profile and immunomodulation in asymptomatic human T-lymphotropic virus type-1 infected blood donors. *J Acquir Immune Defic Syndr*. 2001;27:1–6.
- Tachibana N, Okayama A, Ishizaki J, et al. Suppression of tuberculin skin reaction in healthy HTLV-I carriers from Japan. *Int J Cancer*. 1988;42:829–831.
- Murai K, Tachibana N, Shioiri S, et al. Suppression of delayed-type hypersensitivity to PPD and PHA in elderly HTLV-I carriers. *J Acquir Immune Defic Syndr*. 1990;3:1006–1009.
- Welles SL, Tachibana N, Okayama A, et al. Decreased reactivity to PPD among HTLV-I carriers in relation to virus and hematologic status. *Int J Cancer*. 1994;56:337–340.
- Hisada M, Shima T, Okayama A, et al. Suppression of skin reactivity to purified protein derivative by hepatitis C virus among HTLV-I carriers in Japan. *J Acquir Immune Defic Syndr Hum Retrovirol*. 1998;19:421–425.
- Hisada M, Stuver SO, Okayama A, et al. Gender difference in skin reactivity to purified protein derivative among carriers of HTLV-I in Japan. *J Acquir Immune Defic Syndr*. 1999;22:302–307.
- Suzuki M, Dezzutti CS, Okayama A, et al. Modulation of T-cell responses to a recall antigen in human T-cell leukemia virus type 1-infected individuals. *Clin Diagn Lab Immunol*. 1999;6:713–717.
- Matsuyama W, Mizoguchi A, Iwami F, et al. Clinical investigation of pulmonary Mycobacterium avium complex infection in human T lymphotropic virus type I carriers. *Thorax*. 2000;55:388–392.
- Murphy EL, Wu Y, Ownby HE, et al. Delayed hypersensitivity skin testing to mumps and Candida albicans antigens is normal in middle-age HTLV-I and II infected U.S. cohorts. *AIDS Res Hum Retroviruses*. 2001;17:1273–1277.
- Pape JW, Jean SS, Ho JL, et al. Effect of isoniazid prophylaxis on incidence of active tuberculosis and progression of HIV infection. *Lancet*. 1993;342:268–272.