The Changing Epidemiology of Human T-Cell Lymphotropic Virus Type 1 Infection in Peruvian Female Sex Workers, 1993–2010

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Abstract. Human T-cell lymphotropic virus type 1 (HTLV-1) was the first human retrovirus to be reported and is associated with neoplastic, neurological, autoimmune, and infectious complications. HTLV-1 is endemic in Peru, with the highest prevalence reported among commercial sex workers. Seroprevalence data collected from Peruvian female sex workers (FSWs) working in Callao over three study periods between 1993 and 2010 were used to examine the secular trend in HTLV-1 prevalence. Between 1993 and 2010, the prevalence of HTLV-1 decreased significantly from 14.5% to 3.1% (P < 0.01). The prevalence of HTLV-1 seropositivity differed significantly by birth cohort (1922–1959, 1960–1969, 1970–1979, and 1980–1992), and for each of the four birth cohorts, the prevalence did not significantly decrease by screening year (P > 0.07). There were no cases of HTLV-1 detected among FSW born after 1979 (N = 224). Participant characteristics associated with HTLV-1 seropositivity were birth in the Andes Mountains region, age, increased time in sex work, place of birth, and HIV serostatus, with the odds of HTLV-1 infection decreasing approximately 16% per year (adjusted odds ratio = 0.84, 95% confidence interval = 0.78, 0.90). The increasing use of condoms by later birth cohorts noted in our analysis, as well as the increasing availability of free condoms provided by the Peruvian government—which started in the late 1980s before this study—may have been responsible for declining HTLV seroprevalence.

INTRODUCTION

Human T-cell lymphotropic virus type 1 (HTLV-1) is a positive, single-stranded RNA retrovirus that infects approximately 5–10 million people throughout the world.1,2 Although the majority of people infected with HTLV-1 remain asymptomatic, approximately 5% will develop a neoplastic, neurological, or infectious complication, such as adult T-cell lymphoma/leukemia, HTLV-associated myelopathy/tropical spastic paraparesis, Strongyloides stercoralis hyperinfection, or Norwegian scabies.3,4 Autoimmune complications, such as Sjögren’s syndrome, arthritis, and uveitis, have also been associated with HTLV-1 infection.5 HTLV-1 infection has also been linked with increased risk of acquiring tuberculosis (TB) and has been associated with decreased survival among people coinfected with either TB or human immunodeficiency virus (HIV) seropositivity. The secular trend in declining prevalence persisted after adjustment for age, time in sex work, place of birth, and HIV serostatus, with the odds of HTLV-1 infection decreasing approximately 16% per year (adjusted odds ratio = 0.84, 95% confidence interval = 0.78, 0.90). The increasing use of condoms by later birth cohorts noted in our analysis, as well as the increasing availability of free condoms provided by the Peruvian government—which started in the late 1980s before this study—may have been responsible for declining HTLV seroprevalence.

HTLV-1 infection is endemic in southern Japan, the Caribbean, equatorial Africa, Australia, and South America.1,11,14 Among some populations, more often indigenous, specific human leukocyte antigen (HLA) classes are more likely to have HTLV-1 infection.17,18 In South America, seroprevalence surveys indicate that the ethnic groups with the highest prevalence of HTLV-1 are the Aymara and Quechua-speaking populations living in the Andean Mountains.19 A 1997 study of a Quechua population living in the Andean region of Peru revealed a 5.1% seroprevalence of HTLV-1.20 A cross-sectional study conducted in 2010 among Shipibo–Konoibo women living in the Peruvian Amazon found a 5.9% prevalence of HTLV-1.21–23 In Peru, the prevalence of HTLV-1 has ranged from 0.3% in the general population to 1.3% in blood donors, 1.7% in pregnant women, and 1.8% in men who have sex with men, associated with sexual activity.24–27

Among FSW sampled in Lima, Peru, the overall seroprevalence of HTLV-1 was 7% in 1994 and 3.8% in 1999.13,16 To date, the highest prevalence of HTLV-1 infection in Peru—25%—was reported among female sex workers (FSWs) screened at a public health clinic, Centro de Salud “Alberto Barton” (CSAB) in Callao, Peru, in 1988.28 From 1987 to 1990, a longitudinal study of FSW in Callao found a 1.6% incidence of HTLV-1 infection with 17.6% prevalence on initial screening.29 Commercial sex workers have an increased risk of acquiring sexually transmitted infections (STIs), including HTLV-1, due to occupational risk factors. FSW screened at CSAB between 2008 and 2011 had overall prevalence of 5.1% Chlamydia trachomatis, 0.3% Neisseria gonorrhoeae (NG), 1.7% syphilis, and 1.0% HIV.30

A significant decrease in HTLV-1 prevalence has been reported in Guinea-Bissau (3.5% in 1996 to 2.3% in 2006) and among Haitian women in French Guiana (8.0% in 1991
to 0.3% in 2001.\textsuperscript{31,32} A secular trend of an endemic area, Nagasaki, Japan, noted a significant annual decline in prevalence of HTLV-1 infections, thought to be due to changing demographics, increased condom use, and decreased breastfeeding among later birth cohorts.\textsuperscript{33} Trend analysis of first-time blood donors in Japan has revealed a stable prevalence from 1989 to 1996 (0.47–0.13%) and from 2000 to 2006 (1.05–1.41%).\textsuperscript{34,35} The objective of this analysis was to describe the secular seroprevalence trend and biosociodemographic correlates of HTLV-1 infection in FSW enrolled in a single sexual health clinic in Callao, Peru, during three observational study periods between 1993 and 2010.

METHODS

Study site and population. In Peru, an estimated 15,000 FSW work in the Lima-Callao metro area. Sex work is legally permitted for FSW who are registered and are 18 years of age or older. Registration requires monthly health assessments. The CSAB del Callao is one of two designated STI reference centers to provide health care to registered FSW in Lima, Peru. FSW who receive routine medical care at one of these clinics are considered “registered,” whereas sex workers who do not receive regular medical care, typically women who work in bars or on the streets, are considered “unregistered.” Approximately 5,000 women are registered at the two clinics, and have attended one of the clinics at least once during the past 2 years. Approximately 250 women attend CSAB each month. The number of newly registered FSW averages 700 per year at CSAB.

Study enrollment, procedures, and ethics. FSW 18 years of age or older who presented for medical assessment at CSAB between 1993 and 1997 (Period 1), 2005 and 2006 (Period 2), and 2008 and 2010 (Period 3) were eligible for inclusion. Informed consent was obtained from study participants and the study was approved by the institutional review boards of the University of Washington, Universidad Nacional Mayor de San Marcos, the Directorate of Callao and the U.S. Naval Medical Research Unit No. 6.

For each time period (Period 1: 1993–1997; Period 2: 2005–2006, and Period 3: 2008–2010) each woman completed standardized questionnaires detailing sociodemographic characteristics, sexual practices, and place of work. Participants underwent genital examination with collection of vaginal, endocervical, and blood samples. Genital specimens were tested for NG and Trichomonas vaginalis and blood specimens were tested for syphilis, HIV-1/2, and HTLV-1. Some FSW who enrolled into the study during different time periods underwent repeat serological testing at a subsequent follow-up visit. During Period 2 (2005–2006), Ecuadorian FSW were intentionally oversampled to enable comparisons among FSW characteristics by nationality, but were excluded from this analysis as to focus analysis on Peruvian FSW.

Screening and confirmatory testing for HTLV-1 infection. In each study period, sera were screened for HTLV-1 antibody using enzyme immunoassay (EIA). The name and manufacturer of commercial EIA assays used for each study period were as follows: HTLV EIA, Cambridge Bioscience, Worcester, MA (1993–1997); Vironostika HTLV-I/II Microelisa System, Organon Teknika/Biomerieux, Durham, NC (2005–2006); and Bioelisa HTLV I/II 5.0, BioKit, Barcelona, Spain (2008–2011). Women testing positive for HTLV-1 antibodies by EIA underwent confirmatory testing. Sera from women enrolled in Period 1 (1993–1997) were tested using an rp21e-enhanced Western blot assay (Cambridge Bioscience), with infection defined as immunoreactivity to p24, gp46, and p21env(r) bands. If only other viral-specific bands were present, such as p53 or p19, the individual was considered indeterminate. Women enrolled in Period 2 (2005–2006) received confirmatory testing using HTLV I/II Western Blots (Genelabs Diagnostics, Singapore), with positivity defined according to U.S. Public Health Service criteria: immunoreactivity to both the gag gene product p24 and to an env gene product (gp46 and/or gp61/68). Women enrolled during Period 3 (2008–2010) study period with enzyme-linked immunosorbent assay–positive samples underwent confirmatory testing with a line immunoblot assay (HTLV I/II score; Innogenetics, Ghent, Belgium), which defined positivity as the presence of HTLV-1 IgG antibodies in serum, based on two visible antibody bands that included the gp21-I/-II band, or on three or more bands with the sum of the gp46-I and p19-I band intensity greater than the gp46-II band intensity. The presence of the gp21-I/-II band alone, or a combination of any two bands without a detectable gp21-I/-II band, was considered indeterminate. Participants with indeterminate results were excluded from the analysis (N = 20). The sensitivities and specificities of the initial screening tests used in each study and the sensitivities of the confirmatory tests were similar.\textsuperscript{36–39} The confirmatory test used in Study 2 (by Genelabs Diagnostics) was less specific than the other two (specificity of 50% versus > 92%).\textsuperscript{40–42} All women testing positive for HTLV-1 received counseling regarding potential long-term manifestations of HTLV-1 infection.

Statistical analysis. Descriptive statistics were generated using cross tabulations and $\chi^2$ tests to determine significant differences by HTLV-1 serostatus. Univariate logistic regression was used to determine whether subject characteristics were associated with the odds of HTLV-1 infection. To investigate trends in HTLV-1 prevalence over time, the prevalence of HTLV-1 was calculated for each year that subjects were screened. Calendar year (1993–2010) was included as the independent linear variable in regression models. Our dependent variable was HTLV-1 serostatus. Multivariate logistic regression was used to model the temporal trend in HTLV-1 prevalence in the total study population with subject age, place of birth, time in sex work, and HIV-1 seropositivity were included as a priori confounders in adjusted models. Other variables considered as potential confounders included birth cohort and self-reported condom use. To investigate the sensitivity of our study results to different analytic approaches, we also conducted the adjustment of logistic regression models without initially including any a priori confounders. We further investigated the potential impact of migration on our study results by conducting the same analyses of the trend in HTLV prevalence stratified by region of birth (coast or Andean Mountains). A significance level of 0.05 was used for all hypothesis testing. All analyses were performed using Stata version 13.1 (StataCorp, College Station, TX).

RESULTS

Over the three study periods, 1,938 female commercial sex workers were screened for HTLV-1 infection, with 184 (9.6%) testing positive for HTLV-1; 20 women had indeterminate Western blot results and were excluded from further
An indeterminate result may have represented participants in the process of seroconversion; however, follow-up data were not available. Overall, HTLV-1 serostatus differed significantly by place of birth, age of subject, time in sex work, age of starting sex work, HIV seropositivity, and screening year (Table 1; all \( P \) values \( \leq 0.02 \)).

Condom use increased significantly over time (Figure 1) with a reported prevalence of always using a condom of 240/416 (57.7%) in 1993 compared with 106/125 (84.8%) in 2010 (\( P < 0.01 \)). Although the increase in condom use has been correlated with the decline of HTLV seroprevalence, we found reported condom use within the last week (always/less than always) was more frequent among participants with HTLV-1 infection (70.2% versus 67.3%), although the association was not significant (\( P = 0.43 \)).

The prevalence of HTLV-1 decreased significantly from 14.5% in 1993 to 3.1% in 2010 (\( P < 0.01 \)) (Figure 2). This trend remained significant even after adjustment for age, time in sex work, birthplace in the Andes, and HIV status, with the odds of HTLV-1 infection decreasing approximately 16% per year (adjusted odds ratio [aOR] = 0.84, 95% confidence interval [CI] = 0.78, 0.90). Adjustment of multivariate regression models without the inclusion a priori confounders yielded similar results.

We also examined differences in subject characteristics by study period to investigate whether differences in study populations contributed to changes in HTLV-1 prevalence over time. The percentage of FSW born in the Andean region decreased over time (Figure 1); in 1993, 230/420 (39.5%) of FSW were born in this region compared with 33/146 (22.6%) in 2010. Birthplace in the Andes Mountains was associated with HTLV-1 seropositivity (OR = 2.35, 95% CI = 1.73, 3.20). The association of Andes birthplace with HTLV-1 seropositivity remained significant after adjusting for screening year (OR = 2.07, 95% CI = 1.52, 2.83) (\( P < 0.01 \)).

Women screened during Period 1 (1993–1997) had spent significantly less time in sex work as compared with women screened in Periods 2 (2005–2006) or 3 (2008–2010) (\( P < 0.001 \)). Although the odds of HTLV-1 infection decreased since 1993 among all age groups, the youngest age group screened (18- to 25-year-old women) had the greatest decrease in HTLV-1 seropositivity (aOR = 0.49, 95% CI = 0.34, 0.71). The decreasing trend in HTLV-1 prevalence

### TABLE 1
Characteristics of 1,918 female sex workers by HTLV-1 serostatus in Callao, Peru, 1993–2010.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Total (( N = 1,918 ))</th>
<th>HTLV-1 positive (( N = 184 ))</th>
<th>( P ) value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>18–24</td>
<td>499</td>
<td>21 (11.4)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>25–29</td>
<td>507</td>
<td>27 (14.7)</td>
<td></td>
</tr>
<tr>
<td>30–34</td>
<td>383</td>
<td>44 (23.9)</td>
<td></td>
</tr>
<tr>
<td>35+</td>
<td>527</td>
<td>92 (50.0)</td>
<td></td>
</tr>
<tr>
<td>Period</td>
<td></td>
<td></td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Period 1: 1993–1997</td>
<td>1,477</td>
<td>171 (92.9)</td>
<td>Ref</td>
</tr>
<tr>
<td>Period 2: 2005–2006</td>
<td>62</td>
<td>1 (1.0)</td>
<td>0.04</td>
</tr>
<tr>
<td>Period 3: 2008–2010</td>
<td>379</td>
<td>12 (6.5)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Place of birth</td>
<td></td>
<td></td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Coast</td>
<td>1,193</td>
<td>89 (48.4)</td>
<td>Ref</td>
</tr>
<tr>
<td>Andes</td>
<td>574</td>
<td>88 (47.6)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Amazon</td>
<td>151</td>
<td>7 (3.8)</td>
<td>0.21</td>
</tr>
<tr>
<td>Condom use</td>
<td></td>
<td></td>
<td>0.07</td>
</tr>
<tr>
<td>Always/almost always</td>
<td>1311</td>
<td>130 (9.9)</td>
<td>Ref</td>
</tr>
<tr>
<td>Frequently/sometimes/rarely</td>
<td>422</td>
<td>47 (11.1)</td>
<td>0.47</td>
</tr>
<tr>
<td>Never/almost never</td>
<td>128</td>
<td>4 (3.1)</td>
<td>0.02</td>
</tr>
<tr>
<td>Mean years in sex work</td>
<td>2.7 (5.1)</td>
<td>2.6 (4.8)</td>
<td>0.02</td>
</tr>
<tr>
<td>Mean age in years at starting sex work (( \pm \text{SD} ))</td>
<td>27.9 (7.8)</td>
<td>32.2 (9.5)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>HIV positive</td>
<td>10</td>
<td>4 (2.7)</td>
<td>&lt; 0.001</td>
</tr>
</tbody>
</table>

HIV = human immunodeficiency virus; HTLV-1 = human T-cell lymphotropic virus type 1; SD = standard deviation.
among subjects aged 46 and older did not reach significance ($P = 0.08$).

Across all three periods, increasing age was significantly associated with HTLV-1 prevalence ($P < 0.001$ in all three periods). The prevalence of HTLV-1 seropositivity differed significantly, by birth cohort (1922–1959, 1960–1969, 1970–1979, and 1980–1992) even when adjusted for age (Figure 3). For each of the four birth cohorts, the prevalence did not significantly change by screening year ($P > 0.07$). There were no cases of HTLV-1 detected among FSW born after 1979 ($N = 224$).

**DISCUSSION**

Our analysis demonstrates a dramatic decline in HTLV-1 seroprevalence—from 14.5% to 3.1%—among Peruvian FSWs between 1993 and 2010. This decline is even more impressive considering the HTLV-1 seroprevalence in FSW at the same clinic in Callao was 25% in 1988 and 17.6% in 1990. $^{28}$ Individual level data on FSW tested for HTLV-1 in Callao from 1988 to 1993 were not available for analysis. The decreasing trend in HTLV-1 seropositivity detected in our population of FSW remained significant even after controlling for factors such as age, region of birth, years in sex work, and HIV status. Several sociodemographic, biological, and behavioral characteristics were not measured across the entire study period or were not included in the analysis due to substantial missing data and may have confounded the trend in HTLV-1 seroprevalence we observed; these include migration within Peru and a number of high-risk behaviors.

**Breast-feeding and birth cohort.** Breast-feeding exposure and duration have been associated with increased prevalence of HTLV-1 infections, but breast-feeding history was not collected in these data. $^{11,12,15}$ A possible marker for changes in breast-feeding practice is analysis of HTLV-1 prevalence by birth cohorts. Earlier birth cohorts, especially those born before 1970, revealed much higher prevalence of HTLV-1 compared with later birth cohorts. The strong association with birth cohort may be linked to exposures in life before sex work. A prior longitudinal study in Callao has, however, shown acquisition of HTLV-1 during time as a commercial sex worker suggesting that exposure in adulthood played a role in the early 1990s. $^{29}$ The shorter duration of sex work, coupled with more frequent use of condoms reported in our study may have reduced sexual transmission of HTLV-1 in this population.

**Birthplace and migration.** In 1993, over a third (39.5%) of FSW receiving health services at the CSAB reported a birthplace in the Andes Mountains of Peru, compared with 54.8% from coastal areas and 5.7% from the Amazonian region. Over the screening years, the percentage of FSW born in the Andean region decreased to 25.2% (2010). Birthplace in the Andean region was significantly associated with increased risk of HTLV-1 infection; however, the declining trend in HTLV-1 prevalence over time remained significant even after adjustment for region of birth and among participants stratified by region of birth, which indicates that migration from the Andean region to the coast does not entirely explain the trend. $^{7,19,43}$ Although the exact reason for higher prevalence of HTLV-1 infection among Andean populations is unknown, some researchers have posited that geographic, cultural factors, or HLA type may be responsible. $^{44,45}

**Condom use.** Self-reported condom use among Peruvian FSW working in Callao has continued to steadily increase since 1986. Surveys conducted in Callao in 1986 and 1988 found that 2/135 (1.5%) and 196/636 (30.8%) FSW reported always using a condom, respectively. $^{46,47}$ In our data, there was a significant increase in reported "always using a condom" from 1993 (58%) to 2010 (85%), but this behavior was not associated with HTLV-1 infection. Self-report of always using a condom increased for later birth cohorts ($P < 0.01$). Compared with participants born between 1922 and 1959, participants born between 1980 and 1992 were more than three times as likely to report always using a condom (OR = 3.26, $P < 0.01$).

Given the striking increase in self-reported condom use reported by FSW receiving care at the same clinic, it is plausible that increasing condom use among FSW over the study periods played a role in the decline in HTLV-1 prevalence. Government-purchased condoms were first distributed in the late 1980s, suggesting the decline in HTLV-1 seroprevalence among Peruvian FSW could be a direct result of the Peruvian STI control programs. $^{47,48}$ We believe that the lack of association between condom use and HTLV-1 risk that we
observed in this study is related to the difficulty in evaluating condom use via self-report, which has been documented among other FSWs in Latin America. This unexpected finding highlights the need for higher validity measures of condom use in this and other FSW populations.

**Needle sharing.** Although injection drug use was not reported by any FSW and is generally very rare in Peru, a common behavior among FSW working in Callao during the 1980s and 1990s was use of parenteral injection of antibiotics, vitamins, or steroids purchased outside the medical clinic. Needles used for these injections were sometimes shared needles—which may have contributed to higher HTLV-1 seroprevalence. Although over half of FSW reported receiving such injections in 1986, data regarding the use of injectable antibiotics and frequency of sharing needles to inject penicillin were not collected in later studies or these data. It is plausible that higher HTLV-1 seroprevalence was perpetuated through reuse of nonsterile needles during the 1980s and early 1990s.

**Diagnostic techniques and technology.** The diagnostic techniques and criteria have changed over the last three decades and have been cited as a possible cause of higher rates of HTLV-1 prevalence using earlier criteria and technology. Evidence from validation studies indicates that the sensitivities and specificities of our screening tests were comparable except for the confirmatory test used in Study 2, which was less sensitive as compared with the confirmatory tests of Studies 1 and 3. Because a single HTLV-1 seropositive case was detected during Study 2, false positives resulting from the use of a less specific test did not substantially bias our results.

**CONCLUSIONS**

The prevalence of HTLV-1 infections among Peruvian FSW working in Callao, Peru—the previously one of the most endemic populations in the world—has declined significantly over the last two decades. Increasing age, earlier birth cohort, birth place in the Andes, time of sex work, age at time of starting sex work, and HIV seropositivity were positively associated with HTLV-1 infection in this population. The decrease in HTLV-1 infection over time persisted even after adjustment for these factors. Although similar percentages of FSW with and without HTLV-1 infection reported less than 100% condom usage, the increasing availability of free condoms in the late 1980s makes it plausible that increasing condom use by all women may have been an important factor associated with declining HTLV seroprevalence. Unfortunately, we were not able to examine other specific behavioral factors potentially associated with trends, such as HLA type, exposure, and duration of breastfeeding, or higher risk sexual behavior, such as anal receptive intercourse. The intense internal migration from the rural Andes region to the coastal cities during the 1980s appeared to partially explain the decreasing trend; our data also suggest that changes in early life exposures associated with birth cohort contributed to decreasing HTLV prevalence. We believe that the results of our study would be generalizable to populations in other large cities on the Peruvian coast because they also experienced major immigration from Andean Peruvians during the 1980s. With a growing urban population, it is likely that the decrease in HTLV-1 prevalence we observed in this study would impact national trends. A declining prevalence in the Andean region, as suggested by our study, would compound this. More research is needed to confirm the national representativeness of our results.

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