Prevalence and Incidence of Herpes Simplex Virus Type 2 Infection among Male Zimbabwean Factory Workers

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Stored sera from a cohort of 2397 male factory workers in Harare, Zimbabwe, were screened for herpes simplex virus type 2 (HSV-2)–specific antibodies, to estimate the prevalence and incidence of genital herpes infection and to assess the relation between HSV-2 and human immunodeficiency virus (HIV) acquisition. The prevalence of HSV-2 at enrollment was 39.8%. Correlates of HSV-2 seropositivity were HIV seropositivity, marital status, history of sexually transmitted disease (STD), older age, and higher income. The incidence of HSV-2 seroconversion during follow-up was 6.2/100 person-years. Correlates of HSV-2 seroconversion were enrollment while HIV-positive or seroconversion during follow-up, reported genital ulcer, history of STD, and number of sex partners. No evidence was found that HSV-2 infection was more likely to precede HIV or vice versa. HSV-2 and HIV seropositivity are strong markers for high-risk sexual behavior. Improved interventions targeted to populations in which the incidence of either viral infection is high are needed.

New immunologic assays that use type-specific glycoproteins such as gG1 and gG2 distinguish herpes simplex type 2 (HSV-2) from type 1 (HSV-1) infection and permit better understanding of the epidemiology of genital herpes [1, 2]. Demographic characteristics associated with increased HSV-2 prevalence in the United States include female sex, race, older age, lower education level, and lower income [1–4]. HSV-2 seropositivity is also correlated with markers of sexual exposure, such as lifetime number of partners, years of sexual activity, sexual orientation, and history of other sexually transmitted disease (STD) [3, 5, 6]. A limited number of longitudinal studies in the United States find HSV-2 incidence ranging from 2% over 4 years for college students to 5% over 6 months among STD clinic patients [1].

The prevalence and incidence of HSV-2 in sub-Saharan African populations are of particular concern in light of the potential causal association between genital herpes and transmission of human immunodeficiency virus (HIV) [7–11]. Studies on the African continent show a wide variation in HSV-2 prevalence across diverse populations: 20% among surgical patients in Senegal, 28% among Rwandan military recruits, 36% among patients with genital ulcer disease (GUD) in Uganda, 41% among adults in Congo (Kinshasa), 43% among patients with STDs in Tanzania, 60% among men older than 30 years in rural Tanzania, 68% in rural Uganda, 71% among adults in Congo (Brazzaville), and 96% among prostitutes in Senegal [8, 12–15]. Measures of HSV-2 incidence in sub-Saharan Africa are rare; 1 study in rural Uganda found an incidence of 16.7% over 12 months [15].

Stored sera from a longitudinal study of a factory-based HIV prevention program in Harare, Zimbabwe, provided the opportunity to estimate the prevalence and incidence of HSV-2 infection in a population of urban African men at risk for HIV. We report here the prevalence of HSV-2 infection upon enrollment in the study, the incidence of HSV-2 infection during follow-up, and risk factors for the presence and acquisition of HSV-2 infection. We further assess the temporal relation between HSV-2 and HIV seroconversion.

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Methods

Study population. Stored sera screened for HSV-2–specific antibodies originated from male factory workers enrolled in the Zimbabwe AIDS Prevention Project (ZAPP), a longitudinal study established to determine the prevalence, incidence, and correlates of HIV infection and to evaluate a peer education HIV prevention intervention [16–18]. Subjects were recruited and followed up at 40 factories in greater Harare, Zimbabwe, from March 1993 to June 1997. At enrollment and ~6-month intervals, workers were interviewed on HIV-risk-related behaviors and blood was drawn for HIV testing. Risk factors for HIV acquisition have been described elsewhere [17]. In June 1994, factories were randomized to HIV counseling and testing (control) or HIV counseling and testing plus a peer education program (intervention). Subjects in the present analysis were male factory workers who had baseline and follow-up serologic testing prior to 14 June 1997 and who had sufficient stored sera for HSV-2–specific antibody testing. Of the 3381 subjects ever enrolled in the cohort, 2397 (71%) met these criteria. Age, marital status, education, history of genital ulcer, history of STD, and number of sex partners in the last year did not differ between subjects included in the present analysis and those not included. However, subjects screened for HSV-2 had lower monthly income compared with those not screened (Z$734 vs. Z$948, t test $P < .001$), were less likely to report prostitute contact (18.6% vs. 23.8%, χ^2 test $P = .001$), and had a lower prevalence of HIV at enrollment (16.6% vs. 27.5%, χ^2 test $P < .001$). HIV test results were available to subjects through trained counselors within 2 weeks of each blood draw. However, HSV-2 testing was done after study end and results were not available to subjects.

Laboratory methods. A strip recombinant immunoblot assay (RIBA HSV type 1/type 2 SIA; Chiron, Emeryville, CA) was used to detect and differentiate HSV-1– and HSV-2–specific antibodies [9, 19]. The RIBA type 1/type 2 nitrocellulose strips include recombinant antigen bands from HSV-1 (gG1 and gB1) and HSV-2 (gG2 and gD2) as well as controls for IgG. Antibodies specific for HSV-1 will react with gG1 and gB1 antigen bands but not with the gG2 band. HSV-2 antibodies will react with gG2 and gD2 bands but not with gG1 and gB1 bands. Compared with Western blot, the sensitivity of the RIBA assay is 95.1% for HSV-1 and 98.2% for HSV-2; specificity compared with Western blot is 99.4% for both HSV-1 and HSV-2 [19].

The presence of HIV antibodies was demonstrated by the use of a third-generation enzyme immunoassay (EIA; HIV-1/HIV-2; Abbott Laboratories, Abbott Park, IL). Specimens reactive or indeterminate in the Abbott EIA were retested with a second third-generation EIA (Enzygnost Anti-HIV 1/2 Plus; Behring, Marburg, Germany). Samples were considered HIV antibody positive when positive results were obtained from both EIAs. Indeterminate or conflicting results were resolved by Western blot (HIV Blot 2.2; Diagnostic Biotechnology, Singapore).

Statistical methods. HSV-2 prevalence was estimated as the number of subjects testing HSV-2 seropositive at enrollment divided by the number of enrollment specimens screened. HSV-2 incidence was calculated among subjects testing HSV-2 seronegative at enrollment by dividing the number of seroconversions by the person-years of observation. For subjects remaining HSV-2 seronegative, person-years (py) of observation were calculated as the interval of time from enrollment to the most recent date of follow-up. For subjects seroconverting for HSV-2, person-years were calculated as the interval of time from enrollment to the midpoint between the last HSV-2 seronegative test and the first HSV-2 seropositive test. Ninety-five percent confidence intervals (CIs) for incidence estimates were based on a Poisson distribution.

HSV-2 prevalence and incidence were calculated for subpopulations defined by demographic characteristics and reported risk factors relating to sexual exposure. Correlates of HSV-2 prevalence were identified by use of logistic regression analysis. Correlates of HSV-2 and HIV incidence were assessed by Cox proportional hazards analysis. To determine independent associations with HSV-2 acquisition, we first considered variables separately in a series of bivariate analyses. All variables significant in bivariate analysis at $P < .05$ were entered into a full model; those remaining significant were retained in the final model. The continuous variables of age, income, and number of sex partners were assessed as ordered categories. Odds ratios and hazard ratios were calculated for each stratum separately and for the linear trend across increasing strata. The final multivariate models assess the linear trends across increasing strata.

The temporal sequence of HIV and HSV-2 acquisition was assessed by comparison of the dates of seroconversions for subjects who had serologic evidence of both infections. The number of cases in which HSV-2 seroconversion preceded HIV seroconversion was compared with the number of cases in which HIV seroconversion preceded HSV-2 seroconversion by use of the Wilcoxon matched-pairs sign-rank test. Other statistics were used as indicated in the text.

Results

HSV-2 prevalence at enrollment. Of the 2397 male Zimbabwean factory workers screened, 953 (39.8%) were HSV-2–positive at enrollment in the longitudinal study. History of genital ulcer in the year preceding enrollment was reported by 7.0% of HSV-2 seropositive workers and by 2.4% of HSV-2 seronegative workers.

Table 1 presents HSV-2 prevalence by demographic characteristics and reported risk factors. HSV-2 prevalence increased with increasing age, beginning at 6.5% among workers aged 18–20 years and reaching 62.2% among workers 46 years and older. Of all groups examined, HSV-2 prevalence was highest (76.4%) among workers who were also HIV positive at enrollment. Paradoxically, workers with lower education but higher income had higher prevalence of HSV-2; however, the association of lower education and HSV-2 seropositivity disappeared after we controlled for age and income. In multiple logistic regression analysis (table 2), HSV-2 seropositivity was significantly associated with HIV seropositivity (adjusted odds ratio [OR], 7.2; 95% CI, 5.4–9.4), marital status (divorced OR, 4.1; 95% CI, 2.0–8.6; married, reside apart OR, 3.0; 95% CI, 2.2–4.1; married, reside together OR, 2.9; 95% CI, 2.3–3.9), history of any STD in the previous year (OR, 2.1; 95% CI,

Table 1. Prevalence of herpes simplex type 2 (HSV-2)–specific antibodies among male factory workers at enrollment in a cohort, by demographic characteristics and risk factors, Harare, Zimbabwe, March 1993–June 1997.

Variable	*n*	Number HSV-2 seropositive (prevalence, %)	Unadjusted OR (95% CI)
Total	2397[a]	953 (39.8)	—
Age category in years			
18–20	154	10 (6.5)	Referent[b]
21–25	731	124 (17.0)	2.9 (1.5–5.7)[c]
26–30	433	178 (41.1)	10.1 (5.1–19.6)[c]
31–35	332	186 (56.0)	18.3 (9.3–36.1)[c]
36–40	306	182 (59.5)	21.1 (10.7–41.7)[c]
41–45	200	123 (61.5)	23.0 (11.4–46.4)[c]
⩾46	241	150 (62.2)	23.7 (11.9–47.4)[c]
Marital status			
Single	784	113 (14.4)	Referent
Married, reside together	779	382 (49.0)	5.7 (4.5–7.3)[c]
Married, reside apart	779	423 (54.3)	7.1 (5.5–9.0)[c]
Divorced	45	29 (64.0)	10.8 (5.7–20.5)[c]
Widowed	10	6 (60.0)	8.9 (2.5–32.1)[c]
Education			
Secondary or more	1340	356 (26.6)	Referent
Less than secondary	1053	596 (56.6)	3.6 (3.0–4.3)[c]
Income, by quartiles[d]			
Lowest	604	188 (30.1)	Referent[b]
Lower middle	599	203 (33.9)	1.2 (0.93–1.5)
Upper middle	595	295 (49.6)	2.3 (1.8–2.9)[c]
Highest	599	273 (45.6)	1.9 (1.5–2.5)[c]
Reported genital ulcer in last year			
No	2295	886 (38.6)	Referent
Yes	102	67 (65.7)	3.0 (2.0–4.6)[c]
Reported any STD in last year			
No	2144	809 (37.7)	Referent
Yes	253	144 (56.9)	2.2 (1.7–2.8)[c]
No. of sex partners in last year			
0–1	1417	528 (37.3)	Referent[b]
2–4	825	358 (43.4)	1.3 (1.1–1.5)[e]
⩾5	155	67 (43.2)	1.3 (0.92–1.8)
Reported paying for sex in last year			
No	1954	730 (37.4)	Referent
Yes	443	223 (50.3)	1.7 (1.4–2.1)[c]
Enrolled HIV positive			
No	1999	649 (32.5)	Referent
Yes	398	304 (76.4)	6.7 (5.2–8.6)[c]

NOTE. OR, odds ratio; CI, confidence interval; STD, sexually transmitted disease; HIV, human immunodeficiency virus.

[a] Totals for categories do not always add to 2397 because of missing data.

[b] Logistic regression analysis test for linear trend: age OR/year 1.6 (95% CI, 1.5–1.7), $P < .001$; income OR/quartile 1.3 (95% CI, 1.2–1.4), $P < .001$; sex partners OR/category 1.1 (95 % CI, 1.0–1.1), $P < .01$.

[c] $P < .001$.

[d] Median income was ~Z$500 (US $40)/month.

[e] $P < .01$.

1.5–2.9), older age (OR, 1.4 per age group; 95% CI, 1.3–1.5), and higher income (OR per quartile, 1.1; 95% CI, 1.0–1.2).

HSV-2 incidence during follow-up. The incidence of HSV-2 was 6.2/100 py (95% CI, 5.4–7.0), based on 204 seroconversions among the 1444 men who enrolled seronegative and observed for 3316 py of follow-up. History of genital ulcer during the follow-up period was reported by 15.2% of workers who seroconverted for HSV-2 and by 3.2% of workers who did not seroconvert. Of the 51 workers reporting genital ulcers during the follow-up period, 31 (57.4%) seroconverted for HSV-2.

Table 3 displays HSV-2 incidence by demographic characteristics and reported risk factors. Unlike HSV-2 prevalence, HSV-2 incidence did not differ significantly by age, education, or income. Also in contrast to HSV-2 prevalence, single men did not have a lower incidence of HSV-2 compared with married men. Widowers had the highest incidence of HSV-2 (50.0/100 py); the estimate, however, is based on 2 observed seroconversions (95% CI, 6.1–180.5/100 py). HSV-2 incidence was 23.0/100 py (95% CI, 16.7–31.0) among workers who enrolled HIV-positive and 24.8/100 py (17.4–34.4) among workers who seroconverted for HIV during the follow-up period. Variables independently associated with HSV-2 acquisition in multiple Cox proportional hazard analysis (table 4) were being widowed (adjusted hazard ratio [HR], 5.4; 95% CI, 1.3 ± 22.0), HIV seropositive at baseline (HR, 4.7; 95% CI, 3.3 ± 6.7) or HIV seroconverting during follow-up (HR, 3.9; 95% CI, 2.6 ± 5.8), history of genital ulcer (HR, 2.0; 95% CI, 1.2 ± 3.3), history of any STD (HR, 1.9; 95% CI, 1.3 ± 2.9), and number of sex partners (HR per partner, 1.1; 95% CI, 1.0 ± 1.3). HSV-2 incidence was not independently associated with age, education, income, paying for sex, or working at a factory with a peer education HIV prevention program.

Acquisition of HSV-2 and HIV. Figure 1 displays acquisition of HSV-2 and HIV for 4 groups defined by serostatus at enrollment. Among workers negative for both infections at enrollment, 125 seroconverted for HSV-2 alone, 36 seroconverted for HIV alone, and 36 seroconverted for HSV-2 and HIV during follow-up. Among workers positive for HSV-2 but negative for HIV at enrollment, 52 seroconverted for HIV. Among workers negative for HSV-2 but positive for HIV at enrollment, 43 seroconverted for HSV-2. The incidence of HIV among workers who were HSV-2–positive at enrollment was 3.0/100 py, compared with 2.1/100 py among those who were HSV-2–negative at enrollment ($P = .059$, assuming Poisson distribution). The incidence of HSV-2 among workers HIV-positive at enrollment was 23.0/100 py compared with 5.1/100 py among those HIV-negative at enrollment ($P < .001$, assuming Poisson distribution). Preexisting infection with HSV-2 at enrollment (HR, 3.5;

Table 2. Independent associations with herpes simplex type 2 seropositivity among male factory workers at enrollment in a cohort, Harare, Zimbabwe, March 1993–June 1997.

Variable	Adjusted OR[a]	95% CI
Enrolled HIV positive	7.2	5.4–9.4[b]
Divorced	4.1	2.0–8.6
Married, reside apart	3.0	2.2–4.1
Married, reside together	2.9	2.3–3.9
Reported any STD in last year	2.1	1.5–2.9
Age	1.4[c]	1.3–1.5
Income	1.1[c]	1.0–1.2

NOTE. OR, odds ratio; CI, confidence interval; HIV, human immunodeficiency virus; STD, sexually transmitted disease.

[a] Multiple logistic regression analysis, ORs adjusted for other variables in table, OR per age group and per income quartile.

[b] All P values <.001, except for income, $P = .035$.

[c] OR per group for age and income.

Table 3. Incidence of herpes simplex type 2 (HSV-2) among male factory workers during follow-up in a cohort, by demographic characteristics and risk factors, Harare, Zimbabwe, March 1993–June 1997 ($n = 1444$).

Variable	Person-years	HSV-2 seroconversions	HSV-2 incidence/100 py (95% CI)[a]	Unadjusted hazard ratio (95% CI)
Total	3316	204	6.2 (5.4–7.0)	—
Age category in years				
⩽20	422	26	6.2 (4.0–9.0)	Referent
21–25	1299	84	6.5 (5.2–8.0)	1.1 (0.7–1.7)
26–30	580	30	5.2 (3.5–7.4)	0.9 (0.5–1.5)
31–35	349	28	8.0 (5.3–11.6)	1.3 (0.8–2.3)
36–40	291	19	6.5 (3.9–10.2)	1.1 (0.6–2.0)
41–45	147	8	5.4 (2.4–10.7)	0.9 (0.4–2.0)
⩾46	228	9	4.0 (1.8–7.5)	0.7 (0.3–-1.4)
Marital status				
Single	1462	92	6.3 (5.1–7.7)	Referent
Married, reside together	968	58	6.0 (4.6–7.8)	1.0 (0.7–1.3)
Married, reside apart	846	48	5.7 (4.2–7.5)	0.9 (0.6–1.3)
Divorced	37	4	10.8 (3.0–27.7)	1.8 (0.7–4.8)
Widowed	4	2	50.0 (6.1–180.5)	6.9 (1.7–28.2)[b]
Education				
Less than secondary	1108	67	6.0 (4.7–7.7)	Referent
Secondary or more	2208	137	6.2 (5.2–7.3)	1.0 (0.8–1.4)
Income by quartiles				
Lowest	857	54	6.3 (4.7–8.2)	Referent
Lower middle	857	61	7.1 (5.4–9.1)	1.1 (0.8–1.6)
Upper middle	819	47	5.7 (4.2–7.6)	0.9 (0.6–1.4)
Highest	783	42	5.4 (3.9–7.2)	0.9 (0.6–1.3)
Reported genital ulcer[c]				
No	3212	173	5.4 (4.6–6.3)	Referent
Yes	104	31	29.8 (20.3–42.3)	5.4 (3.7–8.0)[d]
Reported any STD[c]				
No	2967	143	4.8 (4.1–5.7)	Referent
Yes	350	61	17.4 (13.3–22.4)	3.6 (2.7–4.8)[d]
No. of sex partners[e]				
0–1	2724	144	5.3 (4.5–6.2)	Referent
2–4	538	53	9.9 (7.4–12.9)	1.9 (1.4–2.5)[d]
⩾5	55	7	12.7 (5.1–26.2)	1.5 (0.86–2.6)
Reported paying for sex[c]				
No	3002	179	6.0 (5.1–6.9)	Referent
Yes	314	25	8.0 (5.2–11.5)	1.3 (0.9–2.0)
Enrolled HIV-positive				
No	3129	161	5.1 (4.4–6.0)	Referent
Yes	187	43	23.0 (16.7–31.0)	4.4 (3.1–6.2)[d]
HIV seroconverted during follow-up				
No	3171	168	5.3 (4.5–6.2)	Referent
Yes	145	36	24.8 (17.4–34.4)	4.6 (3.2–6.6)[d]
Workplace peer education HIV prevention program				
No	1633	102	6.2 (5.1–7.6)	Referent
Yes	1683	102	6.1 (4.9–7.4)	1.0 (0.7–1.3)

NOTE. py, person-years; CI, confidence interval; STD, sexually transmitted disease; HIV, human immunodeficiency virus.
[a] 95% CI on the basis of Poisson distribution.
[b] Cox proportional hazard analysis, $P < .01$.
[c] Reported during any follow-up period.
[d] $P < .001$.
[e] No. of different sex partners reported during first follow-up interval, linear model hazard ratio per partner, 1.1 (95% CI, 1.0–1.2), $P < .001$.

95% CI, 2.2 ± 5.8) and HSV-2 seroconversion during follow-up (HR, 6.7; 95% CI, 4.2 ± 10.7) were strong predictors of HIV acquisition even after we controlled for age, marital status, education, income, history of genital ulcer, history of STD, number of sex partners, and prostitute contact.

The temporal relation of HSV-2 and HIV acquisition was examined for the 435 workers with serologic evidence of both infections by study end. For 317 workers, the temporal sequence of seroconversions could not be determined because they either enrolled seropositive for both infections ($n = 304$) or were found on the same follow-up visit to have seroconverted for both infections ($n = 13$). Of the remaining 118 dually infected workers, 61 acquired HSV-2 first (52 who enrolled HSV-2–positive plus 9 seroconversions) and 57 acquired HIV first (43 who enrolled HIV-positive plus 14 seroconversions). The data did not provide evidence that 1 infection was more likely to precede the other whether we examined all workers with both infections ($P = .830$, sign-rank test) or only those who were

Table 4. Independent associations with herpes simplex type 2 seroconversion among male factory workers during follow-up in a cohort, Harare, Zimbabwe, March 1993–June 1997.

Variable	Adjusted hazard ratio[a]	95% CI
Widowed	5.4	1.3–22.0[b]
Enrolled HIV positive	4.7	3.3–6.7[c]
HIV seroconverted during follow-up	3.9	2.6–5.8[c]
History of genital ulcer	2.0	1.2–3.3[d]
History of any STD	1.9	1.3–2.9[d]
No. of sex partners[e]	1.1	1.0–1.3[b]

NOTE. CI, confidence interval; HIV, human immunodeficiency virus; STD, sexually transmitted disease.

[a] Multiple Cox proportional hazard analysis, hazard ratios adjusted for all other variables in table.

[b] $P < .05$.

[c] $P < .001$.

[d] $P < .01$.

[e] No. of different sex partners reported during first follow-up interval, hazard ratio per partner.

observed to seroconvert for both infections during follow-up ($P = .880$, sign-rank test).

History of genital ulcer was examined by baseline and follow-up HIV serostatus. Of 304 workers seropositive for both infections at baseline, 41 (13.5%) reported history of genital ulcer in the preceding year, compared with 26 (4.0%) of 649 workers who were HSV-2–positive but HIV-negative ($P < .001$, χ^2 test). Of 36 workers who seroconverted for both HSV-2 and HIV, 13 (36.1%) reported a genital ulcer during follow-up, compared with 18 (14.4%) of 125 who seroconverted for HSV-2 only ($P = .004$, χ^2).

Discussion

The prevalence of HSV-2 infection in this general population of male factory workers was high, 39.8%, but plausible given the wide range of estimates for sub-Saharan Africa. Not surprisingly, a strong association between HSV-2 and HIV seropositivity was found, consistent with cross-sectional seroepidemiologic surveys on the African continent [8, 13, 15]. HSV-2 seropositivity was also associated with ever being married, history of STD, older age, and higher income. The latter finding is in contrast to the United States, where lower income is associated with increased HSV-2 prevalence [2, 4]. A possible explanation is that money may afford increased access to sexual partners, particularly prostitutes, to a greater degree among this urban working population in sub-Saharan Africa.

The observed rate of HSV-2 seroconversion was also high, 6.2/100 py. The estimate is comparable to populations in the United States at high risk for STD, such as multipartnered heterosexual STD clinic patients in Atlanta (5% over 6 months) and gay men in San Francisco (5%/year) [1]. One longitudinal study from the African continent (rural Uganda) estimated HSV-2 incidence among men at 11.8% over 12 months; however, the sample size was relatively small (2 seroconversions among 17 men, 95% CI, 1.4%–42.5%) [15]. Thirty-one (15.2%) of 204 workers seroconverting for HSV-2 gave a history of genital ulcer during follow-up and 31 (57.4%) of 54 workers reporting genital ulcers were observed to seroconvert. The latter finding suggests that HSV-2 is the most common cause of genital ulcers in this population. In contrast to HSV-2 prevalence, the incidence of HSV-2 was surprisingly constant across age, marital status (except widowers), education, and income groups. The difference is likely explained by HSV-2 seropositivity reflecting lifetime risk, whereas HSV-2 seroconversion reflects exposure during the study period. Enrollment while HIV-positive or acquisition of HIV during follow-up remained significantly associated with HSV-2 seroconversion after we controlled for other markers of sexual exposure. A discouraging finding was that the presence of a workplace peer education program, which included on-site availability of free condoms and promotion of their use, was not associated with reduced HSV-2 incidence.

The hypothesis that GUD is causally associated with the acquisition of HIV infection has strong appeal. Genital ulceration may enhance acquisition of HIV by disrupting mucosal membranes, providing an easy portal of entry [20, 21]. Several studies, including the present one, have demonstrated a strong correlation between HIV and HSV-2 infection even after controlling for behaviors likely to lead to both infections [7–11]. Nevertheless, whether HSV-2 infection causes a predisposition to acquisition of HIV is debated [21–24]. Studies cannot completely control for behavior and are vulnerable to biases of self-

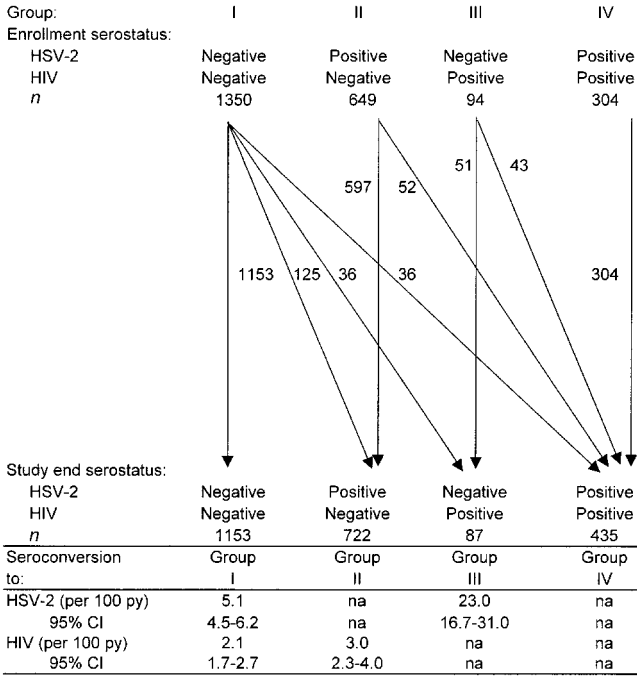

Figure 1. Herpes simplex virus type 2 (HSV-2) and human immunodeficiency virus (HIV) seroconversion in a cohort of male factory workers, Harare, Zimbabwe 1993 to 1997.

reported information. In fact, HSV-2 serology is proposed as a more objective marker for sexual behavior than self-report [5, 12]. Moreover, precise measures of exposure, such as episodes of sexual intercourse with infected partners, are usually not available. Last, many studies were not longitudinal or could not establish whether HSV-2 infection preceded HIV acquisition, a key criterion for causality. At least 2 studies that could determine the temporal sequence of infections found no association between prior HSV-2 infection and HIV seroconversion [21, 24].

The present longitudinal study found that prior HSV-2 infection was associated with a 1.4-fold increase in the rate of HIV acquisition. In contrast, prior HIV infection was associated with a 4.5-fold increase in the rate of HSV-2 acquisition. Moreover, among workers whose seroconversions for both infections were observed during follow-up, more seroconverted for HIV before HSV-2 than vice versa. Overall, however, there was no statistical evidence in this cohort that 1 seroconversion was more likely to precede the other. The evidence would support that either infection predisposes individuals to the other, or, more simply, that persons with either infection are continuing to engage in unsafe sex.

A third explanation is that infection causes increased transmission to others rather than increased susceptibility. Higher incidence of HSV-2 among persons with HIV infection may be observed because people who are HIV positive are more likely to continue to have HIV-positive partners. Because HIV-positive individuals with HSV-2 shed virus more often [25–27], such partners may be more infectious and pose a higher risk of transmission. In this scenario, seropositivity may be a marker for increased infectiousness of sexual partners as opposed to increased susceptibility to infection. Coinfection may enhance transmission of HIV and HSV-2 to others through increased reciprocal viral replication, increased viral shedding, and increased frequency of genital ulceration as a portal for viral excretion. In this cohort, we found a >3-fold increase in reported genital ulcers in the previous year among HSV-2–positive workers who enrolled while HIV positive and a >2-fold increase in genital ulcers among workers who seroconverted for both infections during follow-up. Other studies have demonstrated increased replication of HIV in herpes virus-infected individuals [25, 26] and increased shedding of HSV-2 among HIV-seropositive women compared with seronegative women [27]. Unfortunately, without data on sexual partners in the present study, we cannot draw definitive conclusions.

We recognize other limitations to our data. We were unable to determine the timing of seroconversions for the vast majority of persons with both infections (317 of 435). Precise measures of sexual behavior, sexual exposures, period of active genital lesions, viral replication, viral shedding, and physical examinations were lacking. Moreover, differences between cohort subjects who were included in the study of HIV but not HSV-2 may affect inference on the relation between these infections. Notably, subjects screened for HSV-2 had lower incomes, less prostitute contact, and lower prevalence of HIV.

Whether HSV-2 and HIV transmission are linked through increased susceptibility or through increased infectiousness, the high incidence of both infections in this cohort is evidence of continued high-risk behavior likely to fuel both epidemics. Improved interventions and prevention programs targeted to populations in which the incidence of either infection is high are needed.

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References

1. Stanberrry LR, Jorgensen DM, Nahamias AJ. Herpes simplex viruses 1 and 2. In: Evans AS, Kaslow RA, eds. Viral infections of humans. 4th ed. New York: Plenum Publishing Corporation, **1997**:419–54.
2. Fleming DT, McQuillin GM, Johnson RE, et al. Herpes simplex virus type 2 in the United States, 1976 to 1994. N Engl J Med **1997**; 337:1105–11.
3. Siegal D, Golden E, Washington AE, et al. Prevalence and correlates of herpes simplex infections: the population-based AIDS Multiethnic Neighborhoods study. JAMA **1992**; 268:1702–8.
4. Breinig MK, Kingsley LA, Armstrong JA, Freeman DJ, Ho M. Epidemiology of genital herpes in Pittsburgh: serologic, sexual, and racial correlates of apparent and inapparent herpes simplex infections. J Infect Dis **1990**; 162: 299–305.
5. Cowan FM, Johnson AM, Ashley R, Corey L, Mindel A. Antibody to herpes simplex virus type 2 as a serological marker of sexual lifestyle in populations. BMJ **1994**; 309:1325–8.
6. Wald A, Koutsky L, Ashley RL, Corey L. Genital herpes in a primary care clinic: demographic and sexual correlates of herpes simplex type 2 infections. Sex Transm Dis **1997**; 24:149–55.
7. Holmberg SD, Stewart JA, Gerber AR, et al. Prior herpes simplex virus type 2 infection as a risk factor for HIV infection. JAMA **1988**; 259:1048–50.
8. Langeland N, Haarr L, Mhalu F. Prevalence of HSV-2 antibodies among STD clinic patients in Tanzania. Int J Sex Transm Dis AIDS **1998**; 9: 104–7.
9. Gwanzura L, McFarland W, Alexander D, Burke R-L, Katzenstein D. Association between human immunodeficiency virus and herpes simplex virus type 2 seropositivity among male factory workers in Zimbabwe. J Infect Dis **1998**; 177:481–4.
10. Hook EW III, Cannon RO, Nahamias AJ, et al. Herpes simplex virus infection as a risk factor for human immunodeficiency virus infection in heterosexuals. J Infect Dis **1992**; 165:251–5.
11. Nelson KE, Eiumtrakul S, Celentano D, et al. The association of herpes simplex virus type 2 (HSV-2), Haemophilus ducreyi, and syphilis with HIV infection in young men in northern Thailand. J Acquir Immune Defic Syndr Hum Retroviol **1997**; 16:293–300.
12. Nahmias AJ, Lee FK, Beckman-Nahamias S. Sero-epidemiological and -sociological patterns of herpes simplex virus infection in the world. Scand J Infect Dis **1990**; 69(Suppl):19-36.
13. Kamya M, Nsubuga P, Grant R, Hellman N. The high prevalence of genital herpes among patients with genital ulcer disease in Uganda. Sex Transm Dis **1995**; 22:351–4.
14. Obasi A, Mosha F, Quigley M, et al. Antibody to herpes simplex virus type

2 as a marker of sexual risk behavior in rural Tanzania. J Infect Dis **1999**;179:16–24.

15. Wagner HU, Van-Dyck E, Roggen E, et al. Seroprevalence and incidence of sexually transmitted diseases in a rural Ugandan population. Int J STD AIDS **1994**; 5:332–7.
16. Bassett MT, McFarland W, Ray S, et al. Risk factors for HIV infection at enrollment in an urban male factory cohort in Harare, Zimbabwe. J Acquir Immune Defic Syndr Hum Retroviol **1996**; 13:287–93.
17. Mbizvo M, Machekano R, McFarland W, et al. HIV seroincidence and correlates of seroconversion in a cohort of male factory workers in Harare, Zimbabwe. AIDS **1996**; 10:895–901.
18. Katzenstein D, McFarland W, Mbizvo M, et al. Peer education among factory workers in Zimbabwe: a sustainable HIV prevention intervention. XII International Conference on AIDS. Geneva, June **1998** [abstract 540*/33514].
19. Alexander D, Dinello R, Ashley R, et al. Simultaneous use of derivatives of glycoproteins gB1, gG1, gD2, gG2 as antigens for a type-specific serological assay to diagnose prior infection with HSV-1 or HSV-2. 96th Annual Society for Microbiology General Meeting. New Orleans, LA, May **1996** [abstract C101].
20. Hayes R, Schulz K, Plummer F. The cofactor effect of genita ulcers on the per-exposure risk of HIV transmission in sub-Saharan Africa. Am J Trop Med Hyg **1995**; 98:1–8.
21. Kingsley LA, Armstrong J, Rahman A, Ho M, Rinaldo CR Jr. No association between herpes simplex virus type-2 seropositivity or anogenital lesions and HIV seroconversion among homosexual men. J Acquir Immun Defic Syndr **1990**; 773–9.
22. Cowan FM, Johnson AM, Mindel A. Herpes simplex virus infection as a risk factor for human immunodeficiency virus infection in heterosexuals. J Infect Dis **1993**; 167:772.
23. Rompalo AM, Shepherd M, Lawlor JP, et al. Definitions of genital ulcer disease and variation in risk for prevalent human immunodeficiency virus infection. Sex Transm Dis **1997**; 24:436–42.
24. Keet IPM, Lee FK, van Griensven GJP, Lange JMA, Nahamias AJ, Coutinho RA. Herpes simplex type 2 and other genital ulcerative infections as a risk factor for HIV-1 acquisition. Genitourin Med **1990**; 66:330–3.
25. Heng MC, Heng SY, Allen SG. Co-infection and synergy of human immunodeficiency virus-1 and herpes simplex virus-1. Lancet **1994**; 343: 255–8.
26. Mole L, Ripich S, Margolis D, Holodniy M. The impact of active herpes simplex virus infection on human immunodeficiency virus load. J Infect Dis **1997**; 176:766–70.
27. Augenbraum M, Feldman J, Chirgwin K, et al. Increased genital shedding of herpes simplex virus type 2 in HIV-seropositive women. Ann Intern Med **1995**; 123:845–7.