

Genital Shedding of Herpes Simplex Virus among Men

Anna Wald,^{1,2,4} Judith Zeh,^{3,4} Stacy Selke,⁴ Terri Warren,⁶
Rhoda Ashley,⁴ and Lawrence Corey^{1,4,5}

Departments of ¹Medicine, ²Epidemiology, ³Statistics, and ⁴Laboratory
Medicine, University of Washington, and ⁵Program in Infectious
Diseases, Fred Hutchinson Cancer Research Center, Seattle,
Washington; ⁶Westover Heights Clinic, Portland, Oregon

Epidemiologic studies suggest that most sexual transmission of genital herpes occurs when persons shed virus but lack lesions. This study assessed 79 men (63 with a history of genital herpes simplex virus [HSV] type 2 infection, 5 with a history of genital HSV-1 infection, and 11 with HSV-2 antibodies but no history of genital herpes) and obtained daily swabs for viral culture. HSV was isolated at least once from 60 (81%) HSV-2-seropositive men. The total viral shedding rate in HSV-2-seropositive men was 5%; the subclinical shedding rate was 2.2%. Of 11 HSV-2-seropositive men without a genital herpes history, 7 recognized typical recurrences and HSV was detected in 10. The shedding rate among men with genital HSV-2 was significantly higher than among men with genital HSV-1 infection (odds ratio, 4.4; 95% confidence interval, 1.2–15.3). The frequency of viral shedding in men with genital herpes appears comparable with that in women.

The development of type-specific serologic assays for herpes simplex virus (HSV) infection enables accurate surveys of HSV-2 prevalence in different populations [1]. These studies consistently indicate that HSV-2 infection is more common in women than men, and studies of HSV-2 transmission suggest that women may be more susceptible to acquisition of genital herpes infection than men [2–6]. Potential reasons for greater susceptibility to HSV-2 in women include anatomic differences (e.g., differences in the genital epithelium), longer exposure to inoculum, and a higher rate of viral reactivation among men than women [7]. The frequency and characteristics of subclinical viral shedding among men with genital herpes has received less attention than among women [8, 9]. Here we detail the frequency and sites of viral shedding among men with genital herpes as shown by daily sampling for HSV from the genital area.

Methods

Study population. Two groups of men were recruited for this study. Group 1 included men with a history of genital herpes who were participating in studies of the natural history of genital herpes

at the University of Washington (UW) Virology Research Clinic in Seattle and at the Westover Heights Clinic, Portland, Oregon. Group 2 comprised men with serologically documented HSV-2 infection without a history of genital herpes. This group was recruited from men who tested seropositive for HSV-2 in a survey of patients receiving clinical care at the UW Family Medical Center [10]. The participants without a history of genital herpes underwent a standardized teaching session on genital herpes to help them recognize the clinical manifestations of the infection [11]. Men were eligible to enroll in the study if they were in general good health and not known to be human immunodeficiency virus (HIV) seropositive.

All participants kept a detailed diary of genital signs and symptoms and were seen monthly for clinic visits. The men collected swab samples daily at home for viral cultures. All men obtained swabs of the penile skin and perianal area and 63% obtained an additional sample from the urethral meatus or submitted first morning urine for viral culture as previously reported [12].

Laboratory methods. At study entry, all participants had serologic testing by HSV Western blot [13]. HSV was isolated as previously described and all isolates were confirmed and typed with monoclonal antibodies [14, 15]. Samples obtained from HSV-2-seropositive men with no history of genital herpes were also assayed by HSV DNA polymerase chain reaction (PCR) as described [16].

Statistical analysis. Penile, urethral, urine, rectal, and buttock viral culture samples or lesions were defined as genital. A clinical recurrence was defined as the presence of genital lesions on successive days as noted by the participant; a culture-positive recurrence was one during which virus was isolated from ≥ 1 genital sample. Shedding rates were defined as the number of days on which HSV was isolated from ≥ 1 site, divided by the total number of days on which cultures were obtained, expressed as a percentage. Subclinical shedding rates included only days with no genital lesions in the numerator and the denominator. Episodes of shedding were defined as consecutive days with positive cultures.

Groups of men were compared by the Wilcoxon rank sum test. Risk factors for longer recurrences, shedding, and lesions were

Presented in part: 35th Interscience Conference on Antimicrobial Agents and Chemotherapy, San Francisco, 1995 (abstract H111).

Clinical research followed human experimentation guidelines of the University of Washington Human Subjects Review Committee. All study participants provided written informed consent.

A.M. and T.W. are consultants to GlaxoWellcome (now GlaxoSmithKline).

Grant support: NIH Herpes Program (AI-30731).

Reprints or correspondence: Dr. Anna Wald, University of Washington Virology Research Clinic, 1001 Broadway, Ste. 320, Seattle, WA 98122 (annawald@u.washington.edu).

The Journal of Infectious Diseases 2002;186(Suppl 1):S34–9

© 2002 by the Infectious Diseases Society of America. All rights reserved.
0022-1899/2002/18608S-0004\$15.00

Table 1. Demographic characteristics, history, and type of HSV infection in study participants.

Characteristic	Men with history of genital herpes (n = 68)	Men with HSV-2 antibody without history of genital herpes (n = 11)
Age, median years (range)	35 (21–73)	40 (25–50)
Heterosexual, %	85%	91%
Genital herpes infection, median years (range)	5.8 years (19 days–21 years)	—
Days with cultures, median (range)	61.5 (21–419)	85 (27–305)
Serology		
HSV-1 antibody only	5	0
HSV-2 antibody only	38	4
HSV-1 and -2 antibodies	25	7

assessed via regression or logistic regression, with recurrences or cultures for each man grouped together and adjustments for within-person dependencies and overdispersion [17, 18].

Results

In all, 79 men enrolled in the study and 68 had histories of genital herpes (table 1). The median age was 36 years (range, 21–73); 95% were white. Sixty-eight men were heterosexual and 11 reported having had sex with men (MSM). Seventy-four men were seropositive for HSV-2, 42 for HSV-2 alone, and 32 for HSV-1 and HSV-2. Five men with a history of genital HSV-1 were seropositive for HSV-1 only. Overall, the men obtained viral cultures on 7674 days (median, 64) and we analyzed 26,148 individual site cultures.

Clinical recurrences and viral shedding rates. Among the 74 men with a history of genital HSV-2 infections or HSV-2 antibodies, 57 experienced clinical recurrences of genital herpes during the study period. The mean rate of recurrences was 9 per year; 40 (54%) men had 6 or more recurrences per year. The median duration of recurrences was 5 days (range, 1–22). HSV was isolated from 85 (52%) of 163 recurrences. The culture-positive recurrences were longer than culture-negative recurrences (mean, 7.6 vs. 3.4 days; $P < .001$). Overall, HSV was isolated at least once from 60 men (81%) seropositive for HSV-2. A culture was positive from ≥ 1 site on 343 (5.0%) of 6806 days of culture. The most frequent site of virus isolation was penile skin (265 days), followed by the perianal area (68 days), urine (4 days), and urethra (3 days).

Rates and sites of subclinical shedding in HSV-2-seropositive men. HSV was isolated on days when lesions were absent in 40 men (54%) with HSV-2 antibodies. Overall, of the 343 days on which HSV was isolated, 210 (61%) isolations occurred on days associated with a recurrence and 133 (39%) on days without a recurrence of genital herpes.

The mean subclinical shedding rate was 2.2% on days when cultures were obtained. HSV was not isolated during the study from the genital area in 34 men but was isolated on up to 2% of days in 12 men, on 2%–5% of days in 16 men, and on >5% of days in 12 men (figure 1). The anatomic sites with highest

viral shedding rates were the penile skin (1.4%) and the perianal area (0.8%). Among heterosexual men, the most common site of subclinical shedding was the penile skin, while among MSM, the most common site of shedding was the perianal area.

Figure 2 shows patterns of subclinical viral shedding among men. Subclinical shedding tended to occur on consecutive days. Among 91 episodes of subclinical shedding, 59 (65%) lasted 1 day, 22 (24%) 2 days, and 10 (11%) 3 days. Subclinical viral shedding tended to occur in temporal proximity to clinical recurrences. Among 30 men who had both recurrences and subclinical shedding, the subclinical shedding rate was 3.6%. However, the rate was 10.3% in the week preceding and 3.2% in the week following a clinical recurrence. Thus, 62% of culture-positive days that occurred on days without lesions occurred in the week before or week after a recurrence.

Comparison of men with and without a history of genital herpes. To understand the differences between HSV-2-seropositive men with a history of genital herpes and those without,

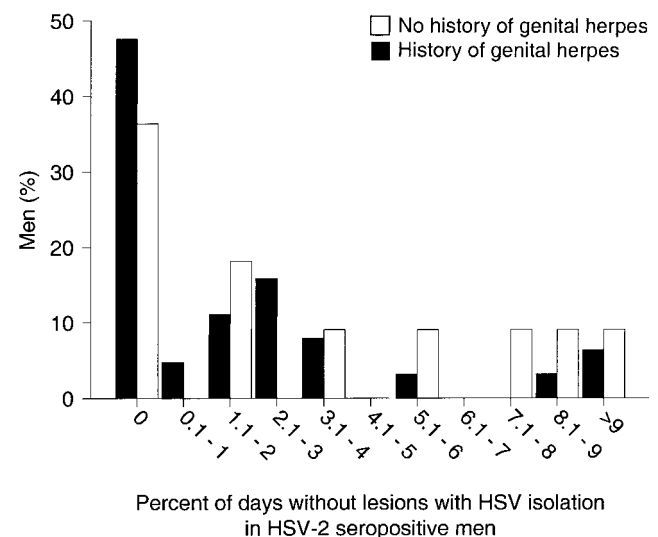


Figure 1. Frequency of subclinical shedding of HSV in the genital tract of HSV-2-seropositive men with and without a history of genital herpes. In all, 63 men had a history of genital herpes, 11 did not.

Subject 6749: HSV-2 Seropositive Heterosexual, History of genital herpes																																
Day	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	
Penile																						+	+	+		+						
Urine/urethral																																
Perianal																																
Lesion(s)																																
Subject 6157: HSV-2 Seropositive Heterosexual, History of genital herpes																																
Day	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	
Penile																																
Urine/urethral																																
Perianal							+								+	+																
Lesion(s)																																
Subject 4835: HSV-2 Seropositive Homosexual, History of genital herpes																																
Day	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	
Penile																								+								
Urine/urethral																																
Perianal		+	+																													
Buttock																																
Lesion(s)																																
Subject 6921: HSV-2 Seropositive Heterosexual, History of genital herpes																																
Day	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	
Penile																																
Urine/urethral			+																													
Perianal																																
Lesion(s)																																
Subject 10103: HSV-1 & 2 Seropositive Heterosexual, No history of genital herpes																																
Day	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	
Penile	+	+									+	+			+	+			+	+							+	+	+	+		
Urine/urethral																																
Perianal																																
Lesion(s)																																

Figure 2. Illustrative diaries of viral shedding and recurrence patterns of 5 men with HSV-2 infection. +, day on which HSV-2 was isolated; shaded area, presence of genital lesions; empty cells, days with negative viral cultures.

we compared the clinical and virologic characteristics of the 2 groups (table 2). Of the 11 HSV-2-seropositive men who entered the study without a history of genital herpes, 7 recognized a typical herpes recurrence during the study. In 4 of these men, HSV was isolated during the recurrence. The mean frequency of recurrences among these men was 5.5 per year and the median duration was 4 days. HSV-2 was isolated in 8 men, resulting in an overall viral shedding rate of 3.6% of days in those without a prior history of genital herpes. The subclinical shedding rate was 3.0% (range, 0–12.7%); 4 of these men shed HSV-2 in the genital tract on >5% of the days without lesions. HSV DNA was detected in 7 of 8 men whose samples were analyzed by PCR, including 2 men whose viral cultures were all negative. Of 667 days with samples, HSV DNA was detected on 79 days (11.8%). Thus, HSV was found in the genital area in 10 of the 11 HSV-2-seropositive men without a history of genital herpes and all 11 either shed virus or recognized a clinical recurrence.

Clinical recurrences and shedding in men with genital HSV-1 infection. Of the 5 men with genital HSV-1 infection, 2 experienced clinical recurrences of genital herpes during the study. The mean rate of recurrences was 1.4 per year and virus was isolated during 25% of recurrences. HSV-1 was isolated in the genital tract on 10 (1.3%) of 789 days in which cultures were

obtained. The most frequent site of virus isolation was the perianal area (10 days) followed by penile skin (2 days) and urine (1 day). Subclinical viral shedding was detected in 3 men with genital HSV-1 infection. Overall, HSV-1 was isolated on 1.2% of days without genital lesions (range, 0–5.5%). The odds ratios for days with lesions, total viral shedding, and subclinical shedding indicated higher rates among HSV-2-seropositive men with a history of genital herpes than for HSV-1-seropositive men with a history of genital herpes. These odds ratios were 14.2 (95% confidence interval [CI], 1.6–125), 4.4 (95% CI, 1.2–15.3), and 1.8 (95% CI, 0.6–5.7), respectively.

Predictors of high subclinical shedding rates. To characterize HSV-2-seropositive men at high risk for subclinical shedding, we modeled potential predictors (table 3). In univariate analyses, time since acquisition of genital herpes and rate of culture-positive recurrences were significantly associated with high subclinical viral shedding rates. Both remained significant in a multivariate analysis.

Discussion

This study shows that in men with genital herpes, reactivation of HSV can occur in the presence or absence of clinical evidence

Table 2. Virologic and clinical characteristics of genital herpes reactivation in HSV-2-seropositive men with and without a history of genital herpes.

Characteristic	History of genital herpes (<i>n</i> = 63)	No history of genital herpes (<i>n</i> = 11)
Shedding rate, days		
Total	302/5680 (5.3)	41/1126 (3.6)
Penile	237/5664 (4.2)	28/1117 (2.5)
Perianal	60/5624 (1.1)	8/1111 (0.7)
Other sites	27/3812 (0.7)	5/1058 (0.5)
Subclinical	100/4912 (2.0)	33/1084 (3.0)
Penile	64/4899 (1.3)	20/1076 (1.9)
Perianal	38/4864 (0.8)	8/1069 (0.7)
Other sites	2/3301 (0.1)	5/1019 (0.5)
Days with lesions, %	13.1%	4.5%
Recurrences		
Rate, mean per year	9.6	5.5
Duration, median days (range)	5 (1–22)	4 (1–11)
Days without lesions as % of all days on which HSV was isolated	100/302 (33)	33/41 (80)

NOTE. Data are no. positive/no. tested (%) unless noted.

of genital lesions. The most common site of subclinical shedding is penile skin of normal appearance. The subclinical shedding rate of 2.2% is similar to the shedding rate in women with genital herpes [8, 9]. As among women, the variability in shedding rates is great and only partly understood. HSV-2 versus HSV-1 infection, short time since acquisition of genital herpes, and high rate of recurrences appear to account for frequent shedding in some men; these same risk factors influence the shedding frequency in women [8]. Of importance, most men without previous clinical evidence of genital herpes but with antibody to HSV-2 also shed virus in the genital tract and experienced previously unrecognized clinical disease.

Until recently, prevention messages about genital herpes focused on abstinence from sexual activity during clinical recurrences. These messages ignore a substantial proportion of days (33%) with viral shedding in the absence of lesions among men with a history of genital herpes and most days (80%) with viral shedding among HSV-2-seropositive men without such history. Yet studies of HSV-2 acquisition suggest that transmission is most likely to occur from partners without a history of genital herpes or on days without recurrences from partners with a history of genital herpes [5, 19].

Our study extends the observations on asymptomatic shedding in men of other investigators. Strand et al. [20] examined urethral shedding in 13 men with genital HSV-2 infection who were followed daily for 4 weeks. HSV was isolated on a single day from 5 men (HSV-1, 4; HSV-2, 1). Straus et al. [21] also investigated asymptomatic shedding among men with genital herpes and found asymptomatic shedding in 8 of 15 participants, but the shedding rates were not reported. In our studies, we defined subclinical shedding as the presence of the virus on days without recognized herpes lesions. Symptoms such as itching were not included as they were found to be nonspecific predictors of viral shedding.

In a recent study of HSV reactivation in MSM, HSV was isolated on 5.5% of days [22]. There were subclinical findings

on 2.7% of days. Most viral shedding in this group was subclinical and the perianal area was the most common site of clinical and subclinical reactivation. These findings parallel those in HIV-infected MSM who had overall higher shedding rates and who shed mostly from the perianal area [23]. Thus, while HIV infection is associated with a high risk of viral shedding, sexual practices also appear to influence the predominant site of HSV shedding. However, heterosexual men still experience viral shedding from the perianal area as the sacral nerves innervate the genital, perianal, and buttocks areas.

We also investigated the natural history of HSV-2 infection in men with serologic but not clinical evidence of infection. Most shed HSV-2 and most recognized recurrences of genital herpes after serologic testing and a teaching session. Serologic surveys consistently show that 75%–90% of HSV-2-infected persons do not know that they have genital herpes [2, 24, 25]. However, after focused education and information about the subject's HSV serostatus, most "asymptomatic" HSV-2-seropositive men recognized recurrences. Among persons with unrecognized HSV-2 infection, 80% of days with viral shedding are not associated with recurrences. This explains why epidemiologic investigations show that persons without a history of genital herpes are a major source of new infections, both to sex partners and to neonates.

Although the initial study of the effect of antiviral therapy on asymptomatic shedding included men and women [21], subsequent studies focused on women [9, 26]. More recently, 27 HSV-2-seropositive men were enrolled in a double-blind randomized crossover study that compared the effects of acyclovir (400 mg twice a day) and valacyclovir (500 mg twice a day) to placebo on subclinical shedding [27]. Participants obtained daily swabs of genital secretions for 3 7-week periods during which they received placebo, acyclovir, or valacyclovir in random order. HSV was isolated on at least 1 day from 24 men during placebo administration, 4 men during valacyclovir therapy, and 5 men during acyclovir therapy. Overall, 3759 days

Table 3. Predictors of high subclinical shedding rates in HSV-2-seropositive men by univariate and adjusted analyses.

Predictor	Odds ratio (95% confidence interval)	
	Univariate	Adjusted
History of genital herpes vs. no history	0.7 (0.3–1.3)	0.5 (0.2–1.0)
MSM vs. heterosexual	1.4 (0.6–3.2)	1.4 (0.6–3.0)
Age/year	0.98 (0.95–1.01)	0.99 (0.96–1.02)
HSV-2 vs. HSV-1 and -2 antibody ^a	1.1 (0.6–2.0)	1.0 (0.5–1.9)
≤1 vs. >1 year since acquisition of genital HSV infection	2.0 (1.1–3.9)	2.0 (1.0–3.7)
Recurrence rate (increase by 1/year)	1.03 (0.99–1.08)	1.00 (0.94–1.07)
Culture-positive recurrence rate (increase by 1/year)	1.08 (1.02–1.15)	1.08 (1.01–1.15)

NOTE. Adjusted analysis for culture-positive recurrence rate is adjusted for time since acquisition. All other adjusted analyses are adjusted for culture-positive recurrence rate (strongest univariate predictor). MSM, men who have sex with men.

^a HSV-2 antibody with vs. without HSV-1 infection.

of sampling were examined, of which 299 (8.0%) were days on which study participants reported lesions. Viral shedding rates decreased with type of therapy from 8.0% on placebo to 0.7% on valacyclovir and 0.6% on acyclovir, 91% and 92% reductions, respectively. When days without lesions were examined separately, the subclinical shedding rate declined from 2.5% on placebo to 0.6% with valacyclovir and 0.7% with acyclovir treatment [27].

Samples were tested for HSV DNA by a PCR assay [16]. HSV DNA was detected in 25 men for ≥1 day during placebo administration, 16 men during valacyclovir therapy, and 17 men during acyclovir therapy. The total viral shedding rate by HSV DNA PCR was 31.9% of days for placebo compared with 8.3% for valacyclovir and 7.1% for acyclovir treatment. The rate of subclinical HSV DNA detection declined from 19.6% during placebo administration to 7.1% during valacyclovir and 6.5% during acyclovir therapy.

Our study showed that men shed HSV-2 at a rate similar to women and that subclinical shedding rates are similar for both sexes. In men, penile skin is the most common site of shedding. As in women, men with recently acquired HSV-2 infection and those with more frequent recurrences are more likely to shed HSV-2 asymptotically. Since rates of infection are consistently higher among women than among men, differences in susceptibility relating to anatomic differences are more likely to play a role in transmission than are reactivation rates. The studies that indicate that antiviral agents are effective in decreasing the rate of asymptomatic shedding in men and women suggest that these agents may decrease sexual transmission of HSV-2. The effect of valacyclovir in reducing the rate of HSV-2 transmission in heterosexual couples is currently under study.

References

- Fleming D, McQuillan G, Johnson R, et al. Herpes simplex virus type 2 in the United States, 1976 to 1994. *N Engl J Med* **1997**;337:1105–11.
- Siegel D, Golden E, Washington AE, et al. Prevalence and correlates of herpes simplex infections: the population-based AIDS in multiethnic neighborhoods study. *JAMA* **1992**;268:1702–8.
- Oliver L, Wald A, Kim M, et al. Seroprevalence of herpes simplex virus infections in a family medicine clinic. *Arch Fam Med* **1995**;4:228–32.
- 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.
- Mertz GJ, Benedetti J, Ashley R, Selke SA, Corey L. Risk factors for the sexual transmission of genital herpes. *Ann Intern Med* **1992**;116:197–202.
- Bryson YJ, Dillon M, Bernstein DI, Radolf J, Zakowski P, Garratty E. Risk of acquisition of genital herpes simplex virus type 2 in sex partners of persons with genital herpes: a prospective couple study. *J Infect Dis* **1993**;167:942–6.
- Benedetti JK, Corey L, Ashley R. Recurrence rates in genital herpes after symptomatic first-episode infection. *Ann Intern Med* **1994**;121:847–54.
- Wald A, Zeh J, Selke S, Ashley RL, Corey L. Virologic characteristics of subclinical and symptomatic genital herpes infections. *N Engl J Med* **1995**;333:770–5.
- Wald A, Zeh J, Barnum G, Davis LG, Corey L. Suppression of subclinical shedding of herpes simplex virus type 2 with acyclovir. *Ann Intern Med* **1996**;124:8–15.
- Wald A, Koutsky L, Ashley R, Corey L. Genital herpes in a primary care clinic: demographic and sexual correlates of herpes simplex type 2 infection. *Sex Transm Dis* **1997**;24:149–55.
- Langenberg A, Benedetti J, Jenkins J, Ashley R, Winter C, Corey L. Development of clinically recognizable genital lesions among women previously identified as having “asymptomatic” HSV-2 infection. *Ann Intern Med* **1989**;110:882–7.
- Wald A, Zeh J, Selke S, et al. Reactivation of genital herpes simplex virus type 2 infection in asymptomatic HSV-2 seropositive persons. *N Engl J Med* **2000**;342:844–50.
- Ashley RL, Militoni J, Lee F, Nahmias A, Corey L. Comparison of Western blot (immunoblot) and G-specific immunodot enzyme assay for detecting antibodies to herpes simplex virus types 1 and 2 in human sera. *J Clin Microbiol* **1988**;26:662–7.
- Langenberg A, Zbanyszczek R, Dragavon J, Ashley R, Corey L. Comparison of diploid fibroblast and rabbit kidney tissue culture and diploid and fibroblast microtiter plate system for the isolation of herpes simplex virus. *J Clin Microbiol* **1988**;26:1772–4.
- Lafferty WE, Krofft S, Remington M, et al. Diagnosis of herpes simplex virus by direct immunofluorescence and viral isolation from samples of external genital lesions in a high prevalence population. *J Clin Microbiol* **1987**;25:323–6.
- Ryncarz AJ, Goddard J, Wald A, Roizman B, Corey L. Development of a high throughput quantitative assay for detecting HSV DNA in clinical samples. *J Clin Microbiol* **1999**;37:1941–7.
- Lipsitz SR, Dear KBG, Zhao L. Jackknife estimates of variance for parameter estimates from estimating equations with applications to clustered survival data. *Biometrics* **1994**;50:842–6.
- McCullagh P, Nelder JA. *Generalized linear models*. New York: Chapman and Hall, **1983**:73.
- Mertz GJ, Schmidt O, Jourden JL, et al. Frequency of acquisition of first-

- episode genital infection with herpes simplex virus from symptomatic and asymptomatic source contacts. *Sex Transm Dis* **1985**;12:33–9.
20. Strand A, Vahline A, Svennerholm B, Wallin J, Lycke E. Asymptomatic virus shedding in men with genital herpes infection. *Scand J Infect Dis* **1986**;18:195–7.
 21. Straus S, Seidlin M, Takiff H, et al. Effect of oral acyclovir treatment on symptomatic and asymptomatic virus shedding in recurrent genital herpes. *Sex Transm Dis* **1989**;16:107–12.
 22. Krone MR, Tabet SR, Paradise M, Wald A, Corey L, Celum CL. Herpes simplex virus shedding among human immunodeficiency virus–negative men who have sex with men: site and frequency of shedding. *J Infect Dis* **1998**;178:978–82.
 23. Schacker T, Zeh J, Hu HL, Hill E, Corey L. Frequency of symptomatic and asymptomatic herpes simplex virus type 2 reactivations among human immunodeficiency virus–infected men. *J Infect Dis* **1998**;178:1616–22.
 24. 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.
 25. Cowan F, Johnson A, Ashley R, Corey L, Mindel A. Relationship between antibodies to herpes simplex virus (HSV) and symptoms of HSV infection. *J Infect Dis* **1996**;174:470–5.
 26. Sacks SL, Hughes A, Rennie B, Boon R. Famciclovir for suppression of asymptomatic and symptomatic recurrent genital herpes shedding: a randomized, double-blind, double dummy, parallel group, placebo-controlled trial [abstract H-73]. In: Programs and abstracts of the 37th Interscience Conference on Antimicrobial Agents and Chemotherapy (Toronto). Washington, DC: American Society for Microbiology, **1997**.
 27. Wald A, Warren T, Hu H, et al. Suppression of subclinical shedding of herpes simplex virus type 2 in the genital tract with valaciclovir [abstract H-82]. In: Program and abstracts of the 38th Interscience Conference on Antimicrobial Agents and Chemotherapy (San Diego). Washington, DC: American Society for Microbiology, **1998**.