Relationship between Antibodies to Herpes Simplex Virus (HSV) and Symptoms of HSV Infection

Frances M. Cowan, Anne M. Johnson, Rhoda Ashley, Lawrence Corey, and Adrian Mindel*

To determine the relationship between antibodies to herpes simplex virus (HSV) types 1 and 2 and diagnosis of orolabial and genital herpes, a cross-sectional survey was done among 869 sexually transmitted disease clinic attenders and 1594 blood donors in London. Among clinic attenders, the prevalence of HSV-1 infection was 59.5% and that of HSV-2 infection was 22.7%, and among blood donors the prevalence was 44.6% and 7.6%, respectively. The sensitivity and specificity of a diagnosis of oral herpes for the presence of HSV-1 antibody was almost identical in the 2 groups (clinic attenders: sensitivity, 33.1%, and specificity, 91.4%; blood donors: sensitivity, 32.3%, and specificity, 94.3%). A diagnosis of genital herpes was less sensitive for antibody for HSV-2 among donors than among clinic attenders (P < .001); however, the specificity was similar in the 2 populations (clinic attenders: sensitivity, 32.1%, and specificity, 96.6%; blood donors: sensitivity, 17.5%, and specificity, 99.5%). False-positive clinical histories were also relatively common (clinic attenders, 12%; donors, 6%). The sensitivity of the diagnosis of genital herpes would be improved if accurate serologic assays for detection of HSV type-specific antibodies were more widely available.

Seroepidemiologic studies in the United States have demonstrated that up to 21.7% of noninstitutionalized adult Americans between 15 and 74 years old are infected with herpes simplex virus (HSV) type 2 [1]. This represents a 32.3% increase in the prevalence of this infection between the late 1970s and late 1980s. Studies that have collected information on clinical history or symptoms due to genital herpes indicate that <=60% of these infections are likely to be unrecognized and hence undiagnosed [2, 3].

In Britain, reports of clinic attendances with genital herpes at sexually transmitted disease (STD) clinics increased 6-fold between 1972 and 1994, when they numbered >24,000 [4]. While these data suggest that the prevalence of genital herpes within the United Kingdom has increased over this period, there may be other explanations. Prior to 1988, no distinction was made between first-episode and recurrent genital herpes, so that a person could appear in the annual statistics more than once. In addition, there may have been more cases diagnosed as a result of increased awareness of the condition over the last 2 decades. Clinic returns only include cases of symptomatic genital herpes diagnosed in STD clinics. We have no knowledge of the extent of diagnosis in other clinical settings or of asymptomatic infection within the general population. Little is known about the extent of clinically apparent orolabial infection except that it is common [5-7].

We report the results of a cross-sectional seroepidemiologic survey of HSV infection among STD clinic attenders and blood donors in London. Herein we describe the relationship between clinical history or symptoms of both orolabial and genital herpes and the presence of antibody to HSV-1 and HSV-2 and discuss the wider implications of such tests in a clinical setting.

Methods

Study population. Eligible patients were all those attending systematically selected routine clinic sessions at a central London STD clinic who had a new clinical problem and who were having a blood sample taken for other reasons. All eligible patients were invited to participate. Study clinics were selected so that each of the three daily clinical sessions was equally represented. Participants completed a structured questionnaire and had sera taken for testing for antibody to HSV. Questions concerned demographic details and sexual history (sexual orientation, age at first sexual intercourse, number of lifetime sex partners, and history of past STDs). Study participants were also asked whether or not they had ever had orolabial or genital herpes diagnosed by a doctor or nurse. Participants with a diagnosis of herpes were asked their age at their first episode. All study participants were asked if they had...
ever had blisters or sores around their mouths or on their genital (penis/scrotum or vulva/vagina) or perianal areas or buttocks and thighs and, if so, with what frequency.

Consecutive blood donors attending a central London donation center were also recruited to the study. After verbal consent, they had a serum sample taken for HSV antibody testing. To maximize the response rate, a shortened version of the STD questionnaire was administered. The questions that appeared in both versions used identical wording except that in the question relating to genital symptoms of herpes, donors were asked only whether they had ever had these symptoms in the genital area. Donors were assured that none of the information collected would be made available to the Blood Transfusion Service.

**Laboratory methods.** Type-specific antibodies to HSV-1 and HSV-2 were detected by Western blot analyses. The laboratory methods have been described in detail elsewhere [8]. Briefly, patient sera were incubated overnight with HSV-1 and HSV-2 cell proteins that had been separated by electrophoresis and transferred onto nitrocellulose strips. Bound antibodies were detected by anti-human antibody and 4-chloronaphthol. Criteria for defining HSV-1-- and HSV-2--specific antibodies are described in detail elsewhere [8]. Each run included four serum controls (HSV-1 antibody-positive, HSV-2 antibody-positive, HSV-1 and HSV-2 antibody-positive, and HSV antibody-negative). Results were expressed as positive or negative for HSV-1 and HSV-2. This assay has been shown to be both sensitive and >99% specific for identifying persons with past HSV-1 or HSV-2 infections or coinfections [8].

**Statistical analyses.** The data were analyzed using SPSS [9] and EGRET [10]. Multiple logistic regression with HSV seropositivity as the dependent variable was used to examine the independent effect of demographic variables, sexual behavior, and past STDs. The results of these analyses have been published [11]. Data on homosexual male STD clinic attenders were not included in this report, as at least 20% were known to be HIV antibody--positive, which is likely to alter the extent to which symptoms are expressed or reported. The relationship between HSV antibody, symptoms, and a diagnosis of herpes was examined ($\chi^2$ for difference in proportions). For the purposes of analysis, a participant was said to have symptoms suggestive of either genital or oral herpes if the person had had one or more episodes of blisters or sores in either the genital or orolabial region.

**Results**

A total of 869 (98%) of 887 clinic attenders and of 1494 (98%) of 1524 blood donors who were eligible to participate agreed to do so. Only data relating to heterosexual clinic attendees were included in these analyses.

**Clinic attenders.** Sufficient sera were available for antibody testing of 347 women and 294 men. The prevalence of antibody to HSV-1 was 55.1% (95% confidence interval [CI], 49.4%--60.8%) for men and 57.6% (95% CI, 52.4%--62.8%) for women; the prevalence of antibody to HSV-2 was 17.3% (95% CI, 13.0%--21.6%) and 24.5% (95% CI, 20.0%--29.3%), respectively. Of clinic attenders tested for HSV antibody, 143 reported a history of orolabial herpes (23.5% men, 22.7% women) and 63 of genital herpes (10.3% men, 10.3% women). Eighty percent of clinic attenders with a history of orolabial herpes had their first episode before age 21. The majority of persons acquired genital herpes between 20 and 40 years, with women acquiring infection earlier than men.

Table 1 illustrates the relationship between a previous diagnosis of orolabial or genital herpes (or both) by HSV antibody status. Of the 282 clinic attenders who had antibody to HSV-1 alone, only 36.2% had had oral or genital herpes (or both) diagnosed in the past. Over 90% (91/101) had had oral rather than genital herpes diagnosed. Similarly, of 63 clinic attenders with antibodies to HSV-2 alone, only 42.8% had a history of oral or genital herpes (or both). The majority (23/27) had genital rather than oral infection. Therefore, among clinic attenders, the positive predictive value of a history of oral herpes for the presence of HSV-1 antibody was 74.8% and that of a history of genital herpes for the presence of HSV-2 antibody was 71.4%. Twenty-five clinic attenders (12.1%) gave a history of herpes but had no serologic evidence of infection. Of these, 19 had a history of oral herpes compared with 7 who had a history of genital herpes ($P < .02$) (1 seronegative participant gave a history of both oral and genital herpes).

**Blood donors.** Serum was available for antibody testing of 639 women and 708 men. The prevalence of antibody to HSV-1 was 43.6% (95% CI, 39.9%--47.3%) among male donors and 45.7% (95% CI, 41.8%--49.6%) among female donors. The prevalence of antibody to HSV-2 was 3.2% (95% CI, 1.9%--4.5%) and 12.4% (95% CI, 9.8%--15.0%), respectively. Of those blood donors who were tested for HSV antibody, 231 had a history of oral herpes (18% men, 23% women), and 25 had had genital herpes (1% men, 3% women). Eighty percent of blood donors with a history of oral herpes had their first episode before they were 21 years old, while 50% of those with a history of genital herpes had their first episode at 21--25 years.

A total of 539 blood donors had antibodies to HSV-1 alone (table 1). Of these, 33.5% had a past diagnosis of herpes; the majority (177/180) had a previous diagnosis of oral herpes. Only 51 blood donors had antibodies to HSV-2 alone. Just 21.6% had a past diagnosis of herpes, the majority (11/12) of whom had genital rather than oral herpes. Among blood donors, the positive predictive value of a history of oral herpes for the presence of HSV-1 antibody was 77.1%; the positive predictive value of a history of genital herpes for the presence of HSV-2 antibody was 48.0%. Forty-two blood donors (6.1%) indicated that they had herpes diagnosed in the past but had no serologic evidence of infection. All but 2 of these (40/42) gave a history of oral rather than genital herpes. Although a similar proportion of clinic attenders and blood donors had a history of oral herpes overall, significantly fewer blood donors reported a history of genital herpes (10.1% vs. 3.3% in blood donors; $P < .001$). Despite the marked differ-
ence in HSV antibody prevalence between the 2 populations, the sensitivity and specificity of a history of oral herpes for the presence of HSV-1 antibody was almost identical in the 2 groups (clinic attenders: sensitivity, 33.1%, specificity, 91.4%; blood donors: sensitivity, 32.3%, specificity, 94.3%). A history of genital herpes identified a significantly lower proportion of those with HSV-2 antibody in blood donors than in clinic attenders ($P < .001$); however, the specificity was similar in the 2 populations (clinic attenders: sensitivity 32.1%, specificity 96.6%; blood donors: sensitivity 17.5%, specificity 99.5%).

**Symptoms suggestive of herpes.** Table 2 illustrates the relationship between HSV antibody and symptoms of herpes among clinic attenders and blood donors. Of those with HSV antibody, blood donors were significantly more likely to be asymptomatic than clinic attenders (clinic attenders, 21.0%; blood donors, 55.7%; $P < .001$). Symptoms suggestive of orolabial herpes were not a sensitive indicator of HSV-1 antibody status (74.5%, clinic attenders; 43.1%, blood donors). Similarly, symptoms of genital herpes were not a sensitive indicator of HSV-2 antibody status (56.2%, clinic attenders; 30.1%, blood donors). The specificity of oral herpes symptoms for HSV-1 antibody was 31.3% among clinic attenders and 89.8% among blood donors; however, the specificity of genital symptoms for HSV-2 antibody was 76.8% among clinic attenders and 96.8% among blood donors.

Clinic attenders were asked about the site of their genital symptoms (genital [vulval/vaginal or penile/scrotal], anal, or buttocks/thighs). Female clinic attenders with HSV-2 antibody were significantly more likely to have genital (odds ratio [OR], 5.5; 95% CI, 3.1-10.1) and anal (OR, 3.3; 95% CI, 1.2-8.8)
### Table 3. History of orolabial and genital herpes, symptoms suggestive of herpes, and HSV antibody prevalence.

<table>
<thead>
<tr>
<th></th>
<th>Antibody to HSV-1 only</th>
<th>HSV-2 only</th>
<th>HSV-1 + HSV-2</th>
<th>Seronegative</th>
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<tr>
<td><strong>Clinic attenders</strong></td>
<td></td>
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<td>History of orolabial herpes</td>
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<td>5 (7.9)</td>
<td>23 (33.8)</td>
<td>19 (9.2)</td>
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<td>Symptoms suggestive of orolabial herpes but no definite diagnosis</td>
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<td>5 (7.9)</td>
<td>11 (16.2)</td>
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<td>Asymptomatic</td>
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<td>53 (84.1)</td>
<td>34 (50.0)</td>
<td>167 (81.4)</td>
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<td>History of genital herpes</td>
<td>11 (3.9)</td>
<td>23 (36.5)</td>
<td>22 (32.4)</td>
<td>7 (3.4)</td>
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<tr>
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<td>14 (22.2)</td>
<td>14 (20.6)</td>
<td>26 (12.7)</td>
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<td>Asymptomatic</td>
<td>227 (80.5)</td>
<td>26 (41.2)</td>
<td>32 (47.1)</td>
<td>172 (83.4)</td>
</tr>
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<td>Total</td>
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<td>63 (100)</td>
<td>68 (100)</td>
<td>205 (100)</td>
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<tr>
<td><strong>Blood donors</strong></td>
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<td>History of orolabial herpes</td>
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<td>15 (27.2)</td>
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<td>2 (3.7)</td>
<td>10 (18.2)</td>
<td>46 (6.4)</td>
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<td>Asymptomatic</td>
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<td>50 (92.6)</td>
<td>30 (54.5)</td>
<td>627 (87.4)</td>
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<td>55 (100)</td>
<td>717 (100)</td>
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<td>7 (12.7)</td>
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<tr>
<td>Symptoms suggestive of genital herpes but no definite diagnosis</td>
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<td>4 (7.2)</td>
<td>10 (18.2)</td>
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<tr>
<td>Asymptomatic</td>
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<td>38 (69.1)</td>
<td>38 (69.1)</td>
<td>687 (96.2)</td>
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<tr>
<td>Total</td>
<td>596 (100)</td>
<td>55 (100)</td>
<td>55 (100)</td>
<td>714 (100)</td>
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</tbody>
</table>

**NOTE.** Data are no. (%).

Symptoms than those without antibody. Men were more likely to have only genital symptoms (OR, 9.7; 95% CI, 5.1-18.3). The presence of HSV-1 antibody was not significantly associated with symptoms at any genital site.

Table 3 shows the relationship between a history and symptoms suggestive of herpes with HSV antibody status. Between 8% and 17% of clinic attenders with serologic evidence of herpes had symptoms of orolabial infection but had not been diagnosed as having herpes. Likewise, 16%–22% of clinic attenders who had symptoms of genital infection had not had genital herpes diagnosed. Among blood donors, 4%–18% of those with HSV antibody had symptoms suggestive of orolabial herpes and 7%–20% had symptoms suggestive of genital herpes, but none had a diagnosis of herpes.

**History of herpes in sex partners of respondents.** Clinic attenders were asked whether any of their sex partners had had oral or genital herpes (a sex partner was someone the respondent had had oral, vaginal, or anal intercourse with). Respondents who answered yes to having a partner with genital herpes had an increased likelihood of having HSV-2 antibody (OR, 6.1; 95% CI, 3.6–10.2), as did those who answered don't know (OR, 2.2; 95% CI, 1.4–3.4) compared with those who answered no. Respondents who answered yes to having a partner with oral herpes had an increased likelihood of having HSV-1 antibody (OR, 2.3; 95% CI, 1.5–3.5).

Of the 89 respondents who had had a sex partner with genital herpes, 35 (39.5%) reported a history of genital herpes themselves; however, of these 35, only 26 had antibodies to HSV-2, 6 had antibodies to HSV-1 alone, and 3 were seronegative. Of the 54 who reported a sex partner with genital herpes and had no history of genital herpes themselves, 16 had antibodies to HSV-2, 26 had antibodies to HSV-1 alone, and 12 were seronegative.

**Discussion**

This is the first study to compare the relationship between a history of herpes, symptoms suggestive of herpes, and HSV antibody prevalence in a low- and high-prevalence population. Overall, only one-third of those with antibodies to HSV had a clinical diagnosis of herpes. The proportion of those with antibodies to HSV-1 who had a clinical diagnosis of oral herpes was broadly similar in both populations. Of note, the proportion of clinic attenders in this study with HSV-2 antibodies who had a diagnosis of genital herpes was comparable to that found in clinic attenders and university students in Seattle despite the lack of similarity between the 3 populations (32%, 30%, and 27%, respectively) [2]. In contrast, the proportion of blood donors with HSV-2 antibodies with diagnosed genital herpes was much less (18%). This is probable because clinic attenders and university students attending for gynecologic examination are more likely to be attending with genital symptoms than donors who were not attending for any medical reason. In addition, some blood donors may have been reluctant to report
a diagnosis of genital herpes while attending the blood donation center despite being reassured that results would not be made available to the Blood Transfusion Service.

Women were more commonly infected with HSV-2 infection than were men (even after controlling for confounding factors) [11], in contrast to HSV-1 infection. This likely reflects the predominant mode of transmission of the two viral types in that HSV-2 is almost always sexually transmitted. Sexual transmission of HSV has been shown to more efficient from men to women than from women to men [12].

Little is known about the prevalence of symptomatic orolabial herpes in the United Kingdom. However, Rawls et al. [13] showed that recurrent orolabial herpes is experienced differently in different populations despite similar levels of infection. The reasons for this were unclear. Findings from this survey indicated that a similar proportion of clinic attenders and blood donors had a clinical diagnosis of orolabial herpes (18%–24%).

Among clinic attenders and blood donors, the site of infection with HSV correlated well with antibody serotype. Among those with HSV-1 antibody, only one-third had a history of orolabial herpes, but only 1.9% had genital herpes. The converse applied in those with HSV-2 antibody alone; 29.4% had a history of genital herpes, but only 6.0% had a history of orolabial herpes. Participants with antibodies to both HSV-1 and HSV-2 were more likely to have a history of both orolabial and genital herpes than those who had antibodies to either type alone.

Several centers in the United Kingdom have reported that ≤50% of cases of genital herpes are due to HSV-1 [5-7]. However, sexual transmission of HSV-1 has generally been considered uncommon [14]. These data show that 3.9% of clinic attenders with HSV-1 antibody had a diagnosis of only genital herpes compared with 0.8% of blood donors. Given that genital HSV-1 infection is less likely to be recurrent and therefore less likely to be diagnosed than genital HSV-2 infection, these data must be regarded as minimum estimates of the extent to which HSV-1 infection is genitally acquired. If only 1 in 2 of those with genital HSV-1 infection is diagnosed as such, these data imply that 8% of HSV-1 infections among clinic attenders may be genitally acquired. There is, however, no way to accurately define the site of infection in a seropositive person on the basis of serologic studies.

Previous studies suggest that orolabial HSV-2 infection is uncommon and recurs infrequently [15]. In this study, 6.0% of persons seropositive for only HSV-2 reported a history of orolabial herpes, which is likely to be a minimum estimate of prevalence. Whether orolabial HSV-2 infection is more common than genital HSV-1 infection is unclear.

Twelve percent of clinic attenders and 6.1% of blood donors had a history of herpes but no serologic evidence of infection. This is likely to reflect misdiagnosis of infections made on clinical grounds alone, as the majority of seronegative, history-positive persons had a clinical diagnosis of orolabial herpes (cold sores), a diagnosis frequently made without confirmatory viral culture.

In this study, a history of the presence of genital blisters or sores (or both) was taken as evidence of symptoms suggestive of genital herpes. However, it is likely that some participants who indicated that they had these symptoms had them for other reasons. Thirteen percent of HSV-seronegative clinic attenders had symptoms suggestive of genital herpes compared with only 4% of blood donors. These data highlight the nonspecific nature of symptoms suggestive of herpes in both populations.

Of note, many more study participants had symptoms suggestive of orolabial or genital herpes than had a history of the infection. While some of these participants have symptoms for reasons other than HSV infection, others may have symptoms but unrecognized herpes. It seems likely that persons who have symptoms of herpes but are unaware of their significance are at particular risk of infecting their sex partners.

This study is the first in the United Kingdom to determine the relative proportions of diagnosed to symptomatic but unrecognized infection. As found in the United States, the majority of HSV infections are frequently asymptomatic and appear to be undiagnosed. Of interest, the group most representative of the general population in London (blood donors) was the group most likely to have unrecognized infection. Even if all those with a history of genital herpes were diagnosed at an STD clinic, this study suggests that the returns to the Department of Health of persons with genital herpes are likely to seriously underestimate the extent of genital herpes and emphasizes the need for further studies to establish the pattern of infection within the United Kingdom. Given the high prevalence of HSV infection within London and the limitations of clinical diagnosis even among physicians trained in the management of STDs, it is important that diagnostic accuracy be improved. Although well-validated type-specific assays are not widely available from commercial laboratories [16, 17], formats amenable to automated performance have recently been reported [18, 19]. These tests should now be evaluated in routine clinical practice to determine whether they enhance the accuracy of diagnosis of HSV infection and improve the management of this infection, particularly among patients who would otherwise have been undiagnosed.

Acknowledgment

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References