

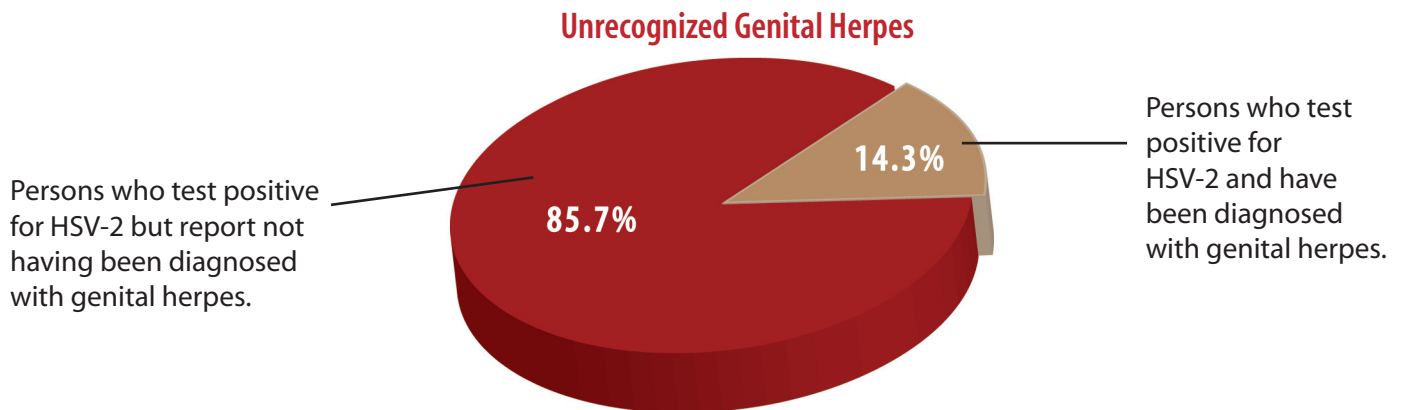
The HERPES Testing Toolkit

A clinician's guide to serologic testing for herpes simplex virus (HSV)

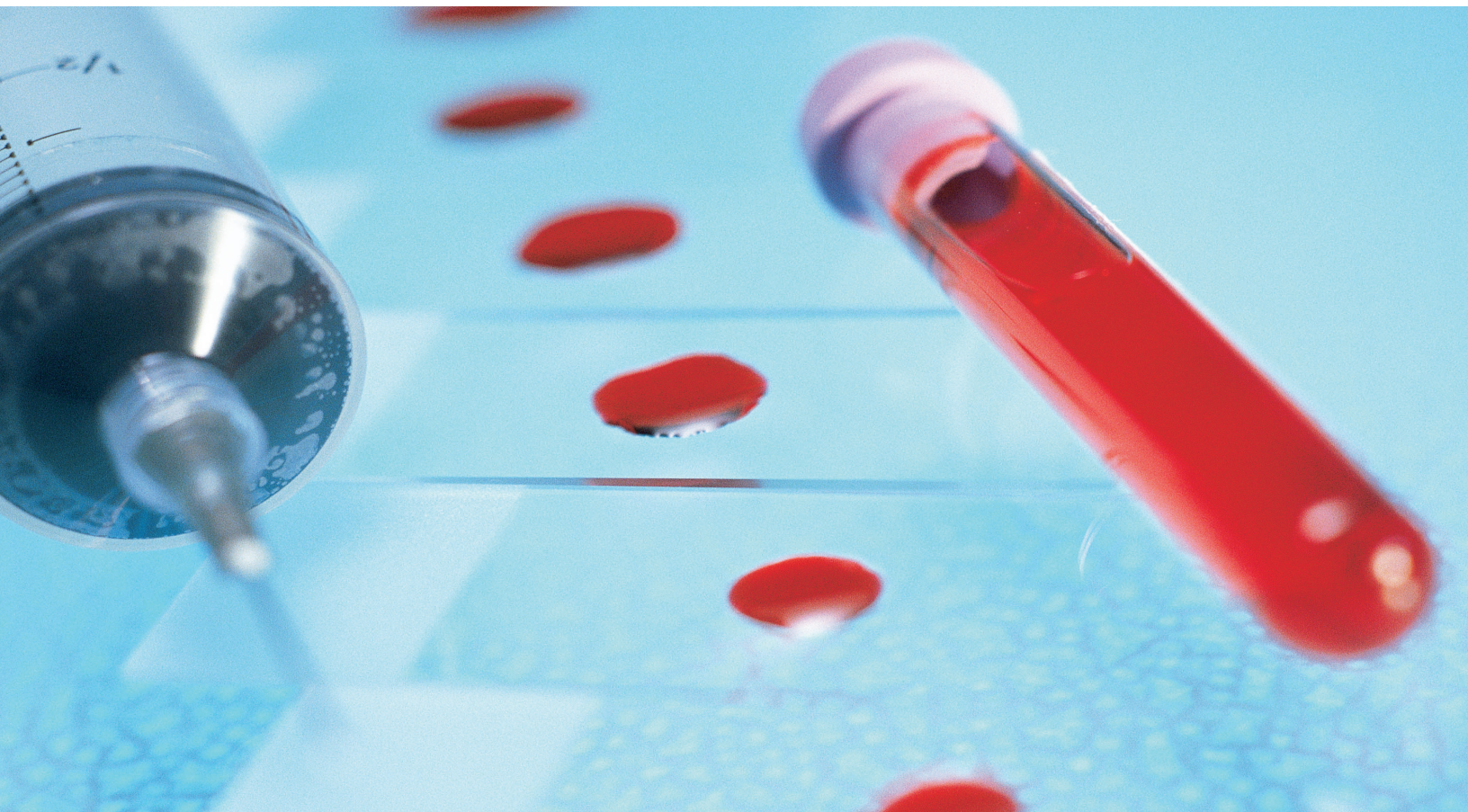
Why is genital herpes testing so important?

Genital herpes is a common viral infection, caused by either herpes simplex virus type 1 (HSV-1) or herpes simplex virus type 2 (HSV-2). It can be easily misdiagnosed and is often underdiagnosed. Clinicians can best serve their patients by using the correct laboratory test to provide a clear diagnosis along with providing education, reassurance, appropriate antiviral therapy, and resources for additional information and emotional support as needed.

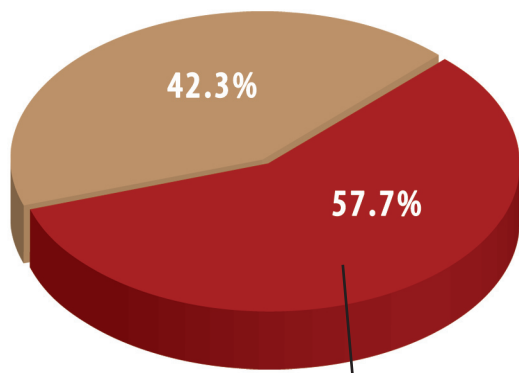
Diagnosing genital herpes by history and clinical examination, without laboratory confirmation, has several serious limitations: 1) 80-90% of people who have genital herpes report no history of signs/symptoms consistent with genital herpes; 2) 20% of people diagnosed by clinical visual exam alone have been found in two studies to not have genital herpes; and, 3) clinical presentations can be subtle, often without genital vesicles and ulcers, leading to misdiagnosis.



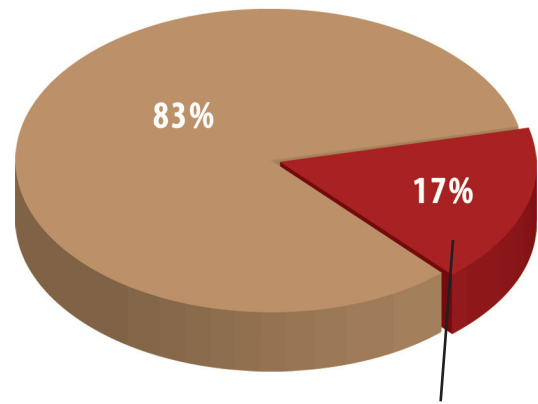
The National Health and Nutrition Examination Survey (NHANES, 1999-2004) shows that only 14.3% of those testing positive for HSV-2 are aware that they have genital herpes.



Seroprevalence of HSV-1 and HSV-2 in the U.S.



Americans seropositive for HSV-1



Americans seropositive for HSV-2

In the United States, more people have genital herpes than all other sexually transmitted infections combined—50 million people in total. Additionally, there are about one million new genital herpes infections each year due to HSV-2. Although the number of cases of genital herpes caused by HSV-1 is difficult to estimate, in some settings, up to half of first clinical outbreaks are due to HSV-1, usually through oral-to-genital transmission. Both HSV-1 and HSV-2 cause life-long infections; however, the natural history of genital infection is substantially different for the two types. Recurrences and asymptomatic viral shedding are much more common with HSV-2. Therefore, determining whether a patient has genital HSV-1 or genital HSV-2 infection is important as it can influence prognosis, treatment, and counseling messages. For example, the suppressive approach to treatment may be more appropriate for those with HSV-2 than for persons with HSV-1.

Who should be tested?

Clinicians should make testing decisions based on individualized risk factors and patient preferences. Although there is some disagreement among experts as to which groups benefit most from serologic (blood) testing, there is general agreement that testing is significantly underutilized. As indicated by the clinical scenarios in the table on page 4, a wide range of patients are likely to benefit from the use of swab and/or type-specific serologic tests (Immunoglobulin G, or IgG, antibody tests).

Swab test options include viral culture and the more sensitive polymerase chain reaction (PCR). These tests are most sensitive when obtained within 3-4 days of the start of an outbreak. Type-specific serologic tests detect the presence of antibodies to HSV, and can be performed in the presence or absence of lesions. Serologic tests cannot determine the anatomic location of HSV disease (i.e., genital versus non-genital). However, there is general agreement that a positive HSV-2 serologic test indicates genital infection with a high degree of certainty. HSV-1 serologic tests are less helpful in determining the site of infection. More than half of the U.S. population has HSV-1 antibodies by late adolescence, primarily due to non-genital infection, thus HSV-1 testing may be less useful than HSV-2 testing. **In general, patients with low or no likelihood of infection should not be tested.** This includes children and those who have not had genital-to-genital or oral-to-genital sexual contact.

CANDIDATE PATIENTS FOR TYPE-SPECIFIC GENITAL HERPES TESTS

Testing Recommended	Swab Test (Viral culture or PCR)	Serologic Test (Type-specific antibodies)
Typical genital lesion	X	X
Clinical diagnosis with negative or no swab test		X
Atypical lesion (e.g. fissure, erythema)	X	X
Recurrent lower genital tract inflammation with no lesion (e.g. dysuria, burning, itching)		X
STI evaluation, no lesion		X
Sexually active patient requests test, no lesion		X
Sex partner of patient with genital herpes		X
Experts disagree on whether or not these patients should be tested		
HIV, no lesion		X
Sexual assault, no lesion		X
Pregnancy, no lesion		X
High risk populations (e.g., men who have sex with men), no lesion		X



What Test Should Be Used?

Many challenges face clinicians regarding the use of genital herpes laboratory tests, especially in the area of serologic test selection and timing of use. The following are important points to consider when selecting a test:

1. Viral detection methods are the best methods for diagnosing genital herpes when lesions are present. However, due to declining sensitivity of viral culture as lesions begin to heal (after 3-4 days), **a negative culture result does not rule out genital herpes**. PCR tests provide a more sensitive alternative for both lesion and skin testing but are not 100% sensitive, can be more costly, and are not readily available in all locations. In either case, to accurately detect virus, the skin must be swabbed vigorously to obtain infected cells and the sample must be placed in the proper collection vial and transported under appropriate conditions. If the swab test is negative or not performed, a serologic test may be the next step.
2. The type-specific serologic tests, while generally accurate, can only detect HSV antibodies after a person's immune response has made detectable antibody. Some patients develop antibodies a few weeks after lesions appear. Others require six weeks, and nearly all will develop antibodies by 16 weeks. The most sensitive tests for detection of early seroconversion include the Focus HerpeSelect® ELISA and biokit HSV-2 Rapid test. By three months, 93% of patients infected with HSV-2 will have seroconverted as measured by HerpeSelect.® **Older, serologic tests that are not type-specific are inaccurate and should never be ordered.**
3. Type-specific serologic tests detect IgG (Immunoglobulin G) antibodies. The current STD Treatment Guidelines from the Centers for Disease Control and Prevention (CDC) state that accurate, type-specific assays for HSV-1 and HSV-2 should be based on HSV type-specific glycoprotein G (gG). **Currently available IgM serologic tests can be falsely positive and should not be used to diagnose genital herpes.**
4. Type-specific serologic tests provide qualitative results. Thus, the exact value does not relate to the severity, recency, or other clinical characteristics of the individual's infection.

Confirmatory and Repeat Testing for Equivocal, Low-value Positive, or Improbable Results

Confirmatory testing is usually not necessary for swab tests or most serologic tests. However, false positive serologic tests occasionally can occur, which can be especially problematic when patients with a low likelihood of HSV infection are tested. Confirmatory serologic tests are indicated in several situations. In early infection, for example, serologic tests may be equivocal or have a low positive value. If persons have an equivocal test because they have only recently acquired herpes and are early in the development of antibodies, the equivocal result may be resolved by repeat testing with a second specimen collected one month after the initial specimen. Alternatively, a second sample can be drawn and sent for a second test using an alternate assay. There are two main confirmatory tests. *Western Blot* is available only at the University of Washington and is useful in confirming HSV-1 and HSV-2 serologic results. *Inhibition assay* for HSV-2 only is available from Focus and Quest Diagnostics and is useful in confirming HSV-2 serologic results. In addition, for low-value and improbable positive results, use of an alternate serologic test (e.g., HerpeSelect®) can be used to confirm the first test.

HSV Serologic Test Comparison

The following table contains summary information on the tests currently most widely used in the U.S. Clinics, hospitals, and other facilities performing HSV tests may find this a useful table for ordering tests and discussing results with patients. Newer type-specific gG-based tests are in development.

Laboratory-based Serology Tests	% Sensitivity (95% CI)*	% Specificity (95% CI)*
HerpeSelect® HSV-1 ELISA**	91 (85-95)	92 (85-97)
HerpeSelect® HSV-2 ELISA**	96 (89-99)	97 (93-99)
HerpeSelect® HSV-1 Immunoblot IgG**	99 (96-100)	95 (89-98)
HerpeSelect® HSV-2 Immunoblot IgG**	97 (91-100)	98 (95-100)
Captia™ HSV-1 ELISA***	88 (82-94)	100 (96-100)
Captia™ HSV-2 ELISA***	97 (89-100)	90 (84-95)
Point-of-Care Serology		
HerpeSelect® Express Rapid HSV-2 IgG**§	98 (95-99) (Capillary whole blood)	99 (97-100) (Capillary whole blood)
biokit HSV-2 Rapid Test§§ Sure-View™ HSV-2 Rapid Test	92 (91-94) (Capillary whole blood)	87 (85-89) (Capillary whole blood)

* Sensitivity and specificity rates vary by population tested. For HerpeSelect®, Captia™, and biokit tests, rates listed are included in the package inserts and are for sexually active adults. Rates are rounded to the nearest whole number.

** A product of Focus Diagnostics.

*** A product of Trinity Biotech.

§ Also FDA cleared for serum and venous specimens.

§§ Also FDA cleared for serum specimens.

Interpreting the Test Results

Swab Viral Tests (Viral Culture and PCR):

Positive: Swab tests have the advantage of identifying both anatomic location of the infection and viral type (though typing often needs to be specifically requested). A genital culture positive for HSV-1, for example, provides definitive diagnosis of genital herpes due to HSV-1. HSV-1 has been increasing as a cause of first episodes of genital herpes. Serologic testing is not necessary in a patient with a recurrent genital lesion who has a prior positive HSV culture that has been typed.

Negative: Swab test results may be negative for many reasons. A negative swab test does not rule out genital herpes. Sensitive swab tests performed early in the next suspected outbreak or type-specific serologic testing may be useful 6-12 weeks after the most recent exposure.

Serologic Tests for HSV-2:

HSV-2 Positive: A positive test for HSV-2 indicates genital infection, as orolabial infection alone is rarely due to HSV-2. Patients who are diagnosed by serologic testing but have no symptoms should be educated about mild, often unrecognized, symptomatic genital herpes disease, asymptomatic viral shedding, and risk of transmission to uninfected sex partners. Confirmatory or repeat testing is usually not necessary, unless, for example, the ELISA value is a low positive in the absence of symptoms. There are various definitions for what constitutes a low positive value, depending on the particular serologic test.

HSV-2 Equivocal: An equivocal result may be resolved by repeat testing with a second specimen collected one month after the initial specimen. If repeating the original serologic test does not resolve the equivocal result, then a confirmatory test should be considered. Rarely, seroconversion can take as long as 6 months after acquisition of infection.

HSV-2 Negative: A negative serologic test for HSV-2 usually means no HSV-2 infection, unless the infection was acquired recently. Because some people can take months to seroconvert, these results should be interpreted cautiously in someone with recent exposure, high-risk behavior, or a new lesion. A repeat test 3-4 months after the initial test may be helpful.

Serologic Tests for HSV-1:

HSV-1 Positive: Slightly more than half of U.S. adults have antibody to HSV-1, so a positive test is not unusual nor does it imply genital herpes, since most people with HSV-1 antibodies have orolabial infection ("cold sores" or "fever blisters"). However, an increasing percentage of new genital herpes infections in young adults appear to be caused by HSV-1. A person with no history of cold sores who has a positive HSV-1 antibody test cannot know for certain where they are infected. These patients should be told that if they develop signs or symptoms in either the oral or genital area, they should come into the office for an evaluation to determine if these are herpetic.

HSV-1 Equivocal: An equivocal result may be resolved by repeat testing with a second specimen collected one month after the initial specimen. If repeating the original serologic test does not resolve the diagnosis, then a confirmatory test should be considered.

HSV-1 Negative: A negative serologic test for HSV-1 usually means no infection with HSV-1 unless it was acquired recently. Of note, the sensitivity of the Focus HSV-1 ELISA is slightly lower than that of the HSV-2 assay, so the ELISA test may miss almost 1 out of 10 people infected with HSV-1. New acquisition of genital HSV-1 infection is of particular concern for pregnant women without HSV-1 antibodies due to high risk of vertical transmission to the neonate.



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The updated Herpes Testing Toolkit is a resource for clinicians to learn more about herpes testing. ASHA's online Toolkit version has comprehensive and updated information about herpes testing. Please visit: www.ashastd.org/herpes/herpes_toolkit for more detailed information than is included in this printed version.