4th Generation HIV Testing and Pregnancy

Routine HIV testing during pregnancy is a key step in the perinatal prevention cascade. However, due to both the pregnancy itself and the urgency with which positive results require intervention to prevent vertical transmission, testing algorithms may be different from the algorithms used outside of pregnancy. In 2006, the CDC developed testing guidelines for pregnancy, and additional updated guidelines are in development. This document was created in response to common questions received at the national Perinatal HIV Hotline. Information is based on expert opinion, taking into consideration pregnancy’s impact on test results and the implications of a positive test on a pregnancy.

My pregnant patient has a positive HIV screening test. What do I do now?

The most commonly used HIV antibody screening tests include 3rd generation enzyme immunoassays (EIAs) which detect IgM and IgG, and 4th generation EIAs which detect IgM, IgG, and the HIV p24 antigen. All lab-based tests are 3rd or 4th generation tests. These screening tests are then confirmed with diagnostic tests, which detect only IgG. The Western Blot, immunofluorescence assay (IFA), and HIV 1/2 differentiation immunoassays (Multispot) are examples of diagnostic tests.

For positive 3rd and 4th generation screening tests, the CDC has proposed confirmatory testing with HIV1/2 differentiation immunoassays (in place of the traditional Western Blot). If there is discrepancy between the 3rd or 4th generation test and the HIV1/2 differentiation immunoassay, the CDC recommends using a viral load (RNA PCR) to rule out acute HIV. For information on these proposed testing algorithms (regardless of pregnancy status), please see: http://www.cdc.gov/hiv/testing/

For positive tests on labor and delivery or in any acute setting, see below for modified recommendations. Please note: a positive rapid test on labor and delivery should always be treated as a true positive while confirmatory results are obtained.

Why is the CDC changing its proposed testing algorithm?

With 3rd and 4th generation testing, there have been multiple cases of false negative results when using traditional confirmatory tests such as Western Blot or immunofluorescence assay (IFA) alone. A positive 3rd or 4th generation test with a negative Western Blot, IFA, or Multispot should be confirmed with an HIV viral load (i.e. RNA PCR) and evaluation for HIV-2. For details of confirmatory testing for HIV, please see: http://www.cdc.gov/hiv/testing/
How is HIV confirmatory testing affected by pregnancy?

While the CDC recommends alternatives to the Western Blot for confirmatory testing, many labs still use this test. Pregnant women who have a positive HIV EIA are more likely to have negative or indeterminate Western Blots. After a positive HIV EIA, the differential diagnosis for an indeterminate or negative Western Blot is acute HIV, HIV-2, or a false-positive EIA result. Acute HIV in pregnancy is associated with increased risk of perinatal transmission. Consequently, diagnosis of acute HIV in a timely manner is essential to allow maximal time for interventions to reduce transmission risk. On the other hand, a false-positive result in pregnancy can lead to unnecessary interventions for a mother and her child. Viral load testing can help differentiate between these results. Please see the section 'How can testing algorithms in pregnancy be altered when expeditious results are required?'

To date, there are limited data on confirmatory testing with HIV1/2 differentiation immunoassays (i.e. the Multispot) in pregnancy. However, this confirmatory testing sequence has obvious benefits: differentiation immunoassays take 15 minutes to run (significantly faster than Western Blots), allowing for rapid confirmation of results. The CDC draft recommendations propose confirmatory testing on labor and delivery for all 3rd and 4th generation positive tests with HIV1/2 differentiation immunoassays.

How do I interpret a negative or indeterminate Western Blot?

The differential diagnosis for a negative or indeterminate Western blot in pregnancy is acute HIV, HIV-2, or a false positive EIA result. Discrepant results may occur because blood samples from pregnant women have increased cross reactivity with the p24 antigen, which is thought to be due to increased circulating auto-antibodies in pregnancy. In addition, in communities where HIV prevalence is low (which is the case for most pregnant women in the US), a positive EIA result is more likely to actually be a false positive. Positive predictive values have been reported as low as 30% in pregnant women in areas of low HIV prevalence. Indeterminate and negative Western blots should be followed-up with a viral load and evaluation for HIV-2. HIV-2, which is endemic in Western Africa but uncommon in the US, can be diagnosed using an HIV 1/2 differentiation immunoassay. Additional confirmatory testing for HIV-2 is available through the CDC.

How does pregnancy affect testing algorithms?

While pregnant women are more likely to have a positive EIA with a negative or indeterminate Western Blot, testing in pregnancy is essential to preventing vertical transmission, and should not be deferred out of concern for having a false positive result. According to the USPHS Perinatal HIV Guidelines, a positive rapid test on labor and delivery should be treated as a true positive while a confirmatory result is obtained. However, outside of an acute setting, a positive EIA should be expeditiously confirmed to

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allow for the initiation of preventative measures to reduce the risk of perinatal transmission or to prevent unnecessary interventions, should the woman go into labor.

**How can testing algorithms in pregnancy be altered when expeditious results are required?**

Viral load (quantitative RNA PCR) testing allows for ruling out acute HIV. While this test is not FDA approved for diagnostic purposes, it is widely available, has a rapid turn-around if done onsite, and undetectable viral load results essentially eliminate the possibility of acute HIV. Viral load testing in conjunction with an indeterminate or negative Western Blot or a positive rapid EIA on labor and delivery can allow for expedited interventions or can prevent unnecessary interventions for pregnant women. For example, if an EIA returns positive on a woman’s first HIV test in pregnancy in the late 2nd or early 3rd trimester, sending a viral load simultaneously with the confirmatory testing will expedite a diagnosis. Also, on labor and delivery, sending a STAT viral load following a positive rapid EIA may change the recommendation for mode of delivery, breast feeding, or the need for neonatal post exposure prophylaxis with antiretrovirals. Rarely, viral load testing will measure low level viremia (<5,000 copies); these are often false positive results. In these situations, expert consultation is required to interpret the results.

**How fast can a lab turn around confirmatory results?**

In emergent situations such as a positive rapid test on labor and delivery, an HIV 1/2 differentiation immunoblot (if available) can be obtained in 15 minutes. If this test is positive, HIV is confirmed. If it is negative, acute HIV must be ruled out with an RNA test (viral load). Viral loads can be reported in approximately 6 hours in some institutions, but require informing the lab director that expedited results will prevent unnecessary interventions for the mother and/or neonate. Providers should consider sending a viral load simultaneously with confirmatory testing in the emergent setting.

**Should a patient with an early false positive test have repeat testing later in pregnancy?**

The CDC recommends repeat HIV testing in the 3rd trimester for “at risk” women, including women who live in high prevalence areas and have high risk for seroconversion in pregnancy (see [http://www.cdc.gov/mmwr/preview/mmwrhtml/rr5514a1.htm](http://www.cdc.gov/mmwr/preview/mmwrhtml/rr5514a1.htm) for the CDC’s definition of “high risk”). Providers may also consider repeat testing in the 3rd trimester for patients with prior false positive tests to potentially avoid a rapid test on labor and delivery necessitating unnecessary interventions for false positive results.

**How do I manage a positive rapid test on labor and delivery?**

A positive rapid test on labor and delivery should be treated as a true positive result until confirmatory testing is done, even if a false positive result is suspected. Providers are recommended to send a STAT viral load along with usual confirmatory testing, and call their lab director to expedite viral load results. The national Perinatal HIV Hotline is also available for consultation 24 hours a day, 7 days a week at 1-888-448-8765 to assist with these challenging cases.
How should a woman be counseled about her preliminary positive test on labor & delivery?

Please see resources for counseling at http://sfaetc.ucsf.edu/resources/rtdl_resource_manual/.
Specifically, see Slide set: HIV counseling and rapid testing in labor.

Where to obtain more information:

Link to CDC testing GL here: http://www.cdc.gov/hiv/testing/

Link to CDC perinatal GL here: http://www.aidsinfo.nih.gov/guidelines
http://hab.hrsa.gov/deliverhivaidscare/clinicalguide11/cg-209_rapid_testing.html

Please call the HIV perinatal hotline with questions. Consultation is available 24 hours a day, 7 days a week at 1-888-448-8765.

Please send questions or feedback to swebber@nccc.ucsf.edu

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