

# Information Package on the Nebraska Public Health Laboratory Screening and Confirmation for the Human Immunodeficiency Virus

Testing for the presence of HIV may be complex in certain clinical situations. The following is intended to provide background information on the laboratory approach to this problem.

## HIV 1 and 2 Antibody Screen

The screening of an individual for HIV is determined in the laboratory by a combination ELISA method, which detects antibodies directed against HIV1 and HIV2. The result of this screen is either negative or positive. Since this is a screening test, a positive result is considered a “presumptive positive” only and requires a second level of testing for confirmation. This second level test is for the detection of HIV 1 Antibody by Western Blot.

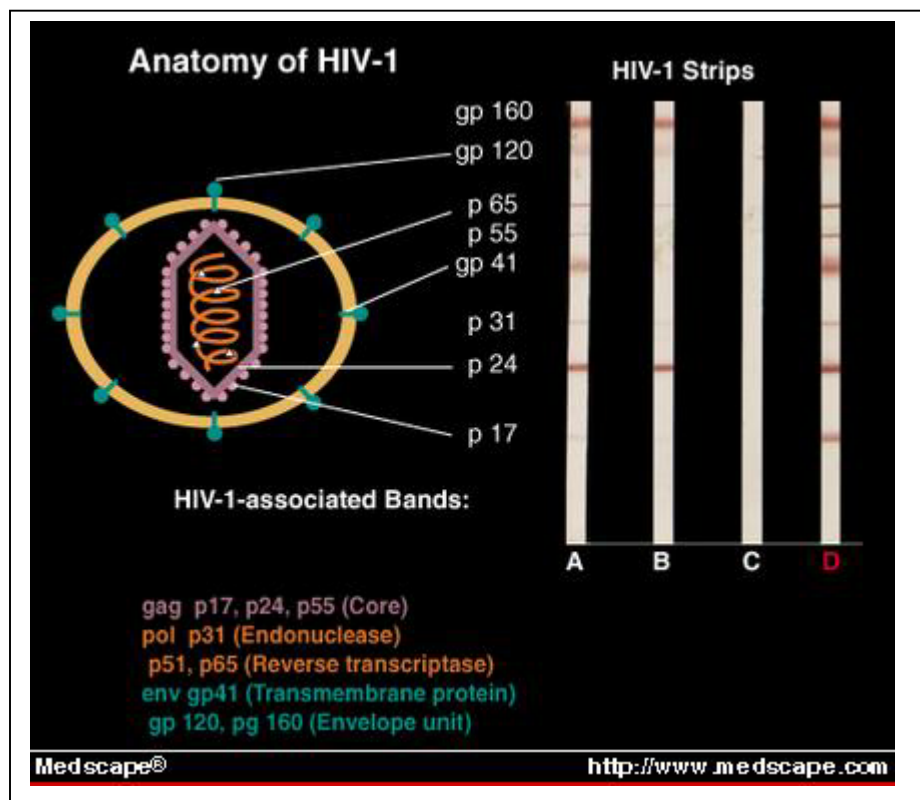


Fig. 1, HIV Western Blot

Please contact Steven Hinrichs, M.D., Director for questions on a specific patient or client.

## **HIV 1 Antibody Detection by Western Blot.**

The interpretation of an HIV 1 Western Blot is made by looking for bands on an immuno blot, a nylon strip showing the presence or absence of bands as shown in figure 1 above. In most situations, two bands need to be present to be positive confirmation test. The bands of importance (named according to their size), are: p 24, gp 40-42, gp 120, or gp160. The confirmation test is interpreted as negative if no bands are seen on the blot. At the NPHL, we also review the ELISA result to make sure it is consistent with the Western blot result.

### **Indeterminate Western Blot**

When only one band, such as p24 is seen, the HIV 1 western blot is interpreted as “indeterminate” which means it is neither negative nor positive. Additionally, an HIV 1 western blot result is sometimes unreadable due to blotchiness of the stain caused by the presence of a non-specific antibody and is interpreted as “indeterminate due to non-specific staining”. The approach to an indeterminate result may vary depending on the clinical situation. In certain cases a molecular based assay is appropriate. (See discussion below)

### **Qualitative HIV 1 DNA Detection**

The HIV1 DNA DetectR™ is used to determine whether a patient has HIV DNA within their white blood cells. The result is reported as positive or negative. Because the report states negative or no HIV DNA detected, the test provides more clarity through the quantitative result and may help resolve previously ambiguous test results.

### **HIV 2 Western Blot**

If the HIV 1-2 antibody screen is positive but the HIV 1 western blot is negative, the serum should be evaluated for presence of antibodies to HIV 2. If there is a positive patient history for foreign travel in Africa or exposure to someone who has traveled in Africa, a HIV 2 Western Blot confirmation may be considered. Confirmation of an indeterminate HIV 2 western blot is complex and requires specific consultation.

## **Quantitative HIV RNA Detection**

A HIV RNA quantitative or viral load determination is ordered as part of a treatment plan for the HIV positive patient. This determination aids in the monitoring of anti-viral therapy, and is obtained shortly after the initial diagnosis of HIV infection is made. As with all quantitative tests, the level of sensitivity is important, some tests are able to detect as little as 50 copies of the virus. The report may indicate the specimen has less than 400 copies of virus, for example.

## **HIV Antibody Specimen Requirements**

### ***Routine HIV Antibody Screening:***

#### **HIV 1 and 2 Antibody Screen**

5.0 mL whole blood in clot or SST tube.

Ship at ambient temperature.

Testing performed at NPHL.

#### **HIV 1 Western Blot**

5.0 mL whole blood in clot of SST tube.

Ship at ambient temperature.

Testing performed at NPHL.

### ***Indeterminate Western Blot Confirmation:***

#### **HIV 1 DNA DetectR™ Qualitative**

5.0 mL EDTA whole blood.

Ship at ambient temperature.

Specimen must arrive at NPHL within 24 hours of collection.

Do not collect on Fridays or Weekends.

Specimen tested at Specialty Laboratories, Santa Monica, California.

### ***Viral Load Testing:***

#### **HIV RNA Quantitation**

5.0 mL EDTA whole blood.

Centrifuge and separate sample from cells within 6 hours of collection.

Transfer plasma to a clean tube and freeze.

Ship frozen EDTA plasma

Testing performed at NPHL.

#### **HIV-1 RNA Quantitation Ultrasensitive**

5.0 mL EDTA whole blood.

Centrifuge and separate sample from cells within 6 hours of collection.

Transfer plasma to a clean tube and freeze.

Ship frozen EDTA plasma

Testing performed at NPHL.

### ***Other Testing:***

#### **HIV 2 Western Blot**

5.0 mL whole blood in clot of SST tube.

Ship at ambient temperature.

Testing performed at ARUP Laboratories, Salt Lake City, Utah.

**HIV-2 Proviral DNA, Qualitative, PCR**

5.0 mL EDTA whole blood.

Ship at ambient temperature.

Testing performed at FOCUS Laboratories, Cypress, California.