

## 4<sup>th</sup> Generation HIV testing

### **Background:**

Effective August 24th, 2020, The Pathology Center laboratory at Methodist Hospital began 4<sup>th</sup> generation HIV testing in house.

Fourth-generation tests simultaneously detect HIV-1 and 2 IgM and IgG antibodies, as well as the p24 antigen. The addition of p24 antigen detection allows fourth-generation assays to detect an acute HIV infection approximately 7-11 days earlier in comparison to antibody only testing.

### **Order:**

Please note that the order has changed to **HIV-1/2 Ag/Ab Combo**

### **Specimen Requirements:**

The specimen requirements are unchanged - 4 mL in SST (Gold top) or serum (Red top) *HIV-1/2 Ag/Ab Combo cannot be added on to tubes which have already had chemistry testing performed, at the recommendation of the reagent vendor.*

### **Test Performed:**

Performed 24/7 and the expected turnaround time (TAT) is 3 hours. The expected TAT for the confirmatory testing, if needed, is 5 days.

### **Confirmatory Algorithm:**

Following a repeatedly reactive HIV screening assay, an HIV differentiation assay will be performed as confirmation, per the 2014 CDC algorithm. The HIV 1/2 antibody differentiation assay will be *automatically* performed following a repeatedly reactive HIV antibody screening test. It is not necessary to order the antibody differentiation assay separately.

If the reflex HIV differentiation assay results are negative for HIV-1/2, the overall interpretation of the testing is "**indeterminate**". If the interpretation of the HIV differentiation assay is indeterminate for HIV-1, an HIV-1 by quantitative PCR is recommended. The quantitative PCR is *not* performed automatically and a second sample (EDTA plasma) must be collected.

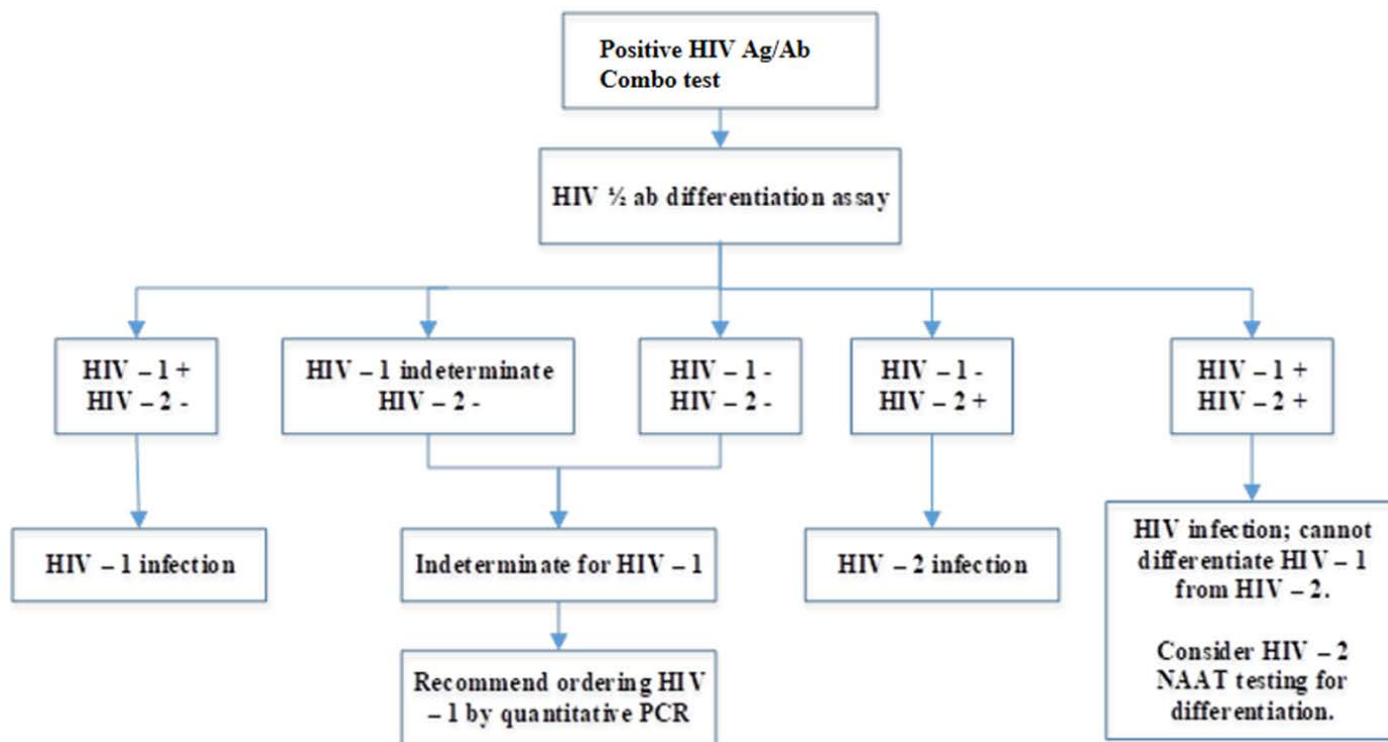
## **Reporting:**

### Initial Ag/Ab in house testing:

- Negative test is reported as "Non-Reactive"
- Positive test is repeated in a duplicate and any positive result on the repeat test is reported as "Reactive" with the following comment: *"Preliminary Reactive Result. Confirmatory Test to Follow"*

### Differentiation Confirmatory testing (Reference laboratory):

- HIV-1 or HIV-2 **Positive** with an interpretive comment:  
*"The specimen is positive for HIV-1 (or HIV-2) antibodies by the HIV-1/HIV-2 antibody differentiation assay.  
If this positive result is inconsistent with additional testing (HIV RNA and CD4 count), testing a new specimen for HIV antibodies would be useful to rule out contamination and mislabeling."*
- Both HIV-1 and HIV-2 **Negative** with an interpretive comment:  
*"HIV Serologic Interpretation - Indeterminate  
HIV nucleic acid testing on a new specimen is recommended.  
The specimen is negative for both HIV-1 and HIV-2 antibodies by the HIV-1/HIV-2 antibody differentiation assay."*



Please direct any questions to Dr. George Bedrnicek at 402 955-5528, Dr. Tess Karre at 402 354-7842 or to Dr. Deborah Perry at 402 354-4559.