Step 1: HIV-1/2 Antigen/Antibody Immunoassay

- In the case of a nonreactive result, the clinician should discuss goal-oriented, harm-reduction strategies, including PrEP and emergency PEP, with any patient who reports recent or likely ongoing HIV risk exposures or refer the patient for prevention services. (A3)
- Clinicians should offer repeat HIV testing every 3 months, or sooner if acute HIV is suspected, for as long as an individual remains at high risk of HIV exposure. (A3)

Step 2: HIV-1/HIV-2 Antibody Differentiation Immunoassay

- Per the standard HIV laboratory testing algorithm, if a reactive result is obtained with an HIV-1/2 Ag/Ab immunoassay testing (step 1), clinicians should perform supplemental testing (step 2) with an FDA-approved HIV-1/HIV-2 Ab differentiation immunoassay. (A1)
  - If the result of the HIV Ab differentiation immunoassay (step 2) is positive for HIV-1 or HIV-2 Abs, the clinician should provide or refer the patient for rapid ART initiation and transmission prevention counseling. (A1)
  - Refer to the NYSDOH Al guideline Rapid ART Initiation.
  - Note: If the HIV Ab differentiation assay result is positive but undifferentiated (i.e., reactive for both HIV-1 and HIV-2), repeat testing may determine if the patient has HIV-1 or HIV-2 infection.

Step 3: HIV-1 Nucleic Acid Testing (qualitative or quantitative HIV RNA testing)

- If the HIV-1/2 Ab differentiation immunoassay (step 2) result is nonreactive (negative), indeterminate (neither positive nor negative for HIV-1 or HIV-2), and the lab does not perform reflex testing, the clinician should immediately order HIV-1 RNA NAT (step 3) to detect the presence of HIV-1 RNA and confirm or exclude HIV-1 infection. (A*)
  - If HIV-1 RNA is detected, the clinician should inform the patient of the acute HIV-1 diagnosis, recommend ART initiation, and prioritize counseling to prevent HIV transmission. (A1)

HIV-1/2 RNA Nucleic Acid Testing (Step 3)
FIGURE 2: HIV Laboratory Testing Algorithm

Step 1: Perform HIV-1/2 antigen/antibody (Ag/Ab) immunoassay

Abbreviations:
Ab, antibody; Ag, antigen; APHL, Association of Public Health Laboratories; CDC, Centers for Disease Control and Prevention; ind, indeterminate; FDA, U.S. Food and Drug Administration; NAT, nucleic acid test; NYSDOH, New York State Department of Health; PEP, post-exposure prophylaxis; PrEP, pre-exposure prophylaxis.

Notes:
a. Adapted from CDC 2018 Quick reference guide: Recommended laboratory HIV testing algorithm for serum or plasma specimens and APHL Suggested reporting language for the HIV laboratory diagnostic testing algorithm.
b. APHL and CDC continue to recommend that laboratories use an FDA-approved instrumented HIV-1/HIV-2 Ag/Ab immunoassay as the initial assay in the laboratory HIV testing algorithm for serum or plasma due to their superior sensitivity for detecting acute HIV infection. However, the FDA-approved single-use rapid HIV-1/HIV-2 Ag/Ab immunoassay may be used as the initial assay in the laboratory HIV testing algorithm for serum or plasma if an instrumented assay is not available.
c. Become familiar with the laboratory's internal testing algorithm and results-reporting policies. Many labs will reflex additional screening steps (such as HIV Ab differentiation immunoassay and HIV RNA) on the original sample without supplemental orders. Other labs may require additional samples or appointments.
d. This includes specimens reported as HIV-2 positive with HIV-1 cross-reactivity.
e. Further testing may be performed to determine type.
f. Per the Geenius package insert, specimens with this final assay interpretation should be retested with a new cartridge. If the final assay interpretation is again HIV-2 indeterminate, it should be reported as such and followed with an HIV-1 NAT.
g. Most laboratories reflex directly to an HIV-1 RNA test without requiring an additional test order or new specimen, either by performing the test in-house or referring the specimen to another laboratory. If the laboratory is unable to or does not automatically reflex directly to the RNA test, clinicians should order an HIV-1 RNA test as soon as possible. To order directly to an HIV-1 NAT, test 1st to confirm presence of HIV-1 RNA. If HIV-1 RNA is detected, proceed with testing for HIV-2.
h. A negative HIV-1 NAT result and repeatedly HIV-2 indeterminate or HIV indeterminate antibody differentiation immunoassay result should be referred for testing with a different validated supplemental HIV-2 test (antibody test or NAT) if available. Alternatively, redraw and repeat algorithm in 2 to 4 weeks to assess HIV-2 infection.

HIV antibodies detected:
HIV antibodies NOT detected or results are indeterminate (ind):
HIV antibodies detected:
HIV negative
HIV-1 (-)
HIV-2 (-)

HIV RNA is not detected. Patient is negative for acute HIV-1 infection.

HIV antibodies detected:
HIV-1 Ab (+)
HIV-2 Ab (-)

Step 2: Perform HIV-1/HIV-2 Ab differentiation immunoassay

Most labs will perform this as a reflex test when the initial test is reactive.

HIV antibodies detected:
HIV-1 Ab (+)
HIV-2 Ab (+)

Step 3: Perform HIV-1 nucleic acid testing (NAT)

Most labs will perform this as a reflex test if the differentiation assay is positive.