

HIV TESTING ALGORITHMS

A STATUS REPORT

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Executive Summary

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Executive Summary

HIV Testing Algorithms: A Status Report

In 1989 the Centers for Disease Control and Prevention (CDC) and the Association of Public Health Laboratories (APHL) released recommendations for a sequential, two-test algorithm for serodiagnosis of HIV-1 infection. In this algorithm, screening is conducted with an enzyme immunoassay (EIA). Specimens repeatedly reactive by EIA are subjected to a more specific supplemental test, either a Western blot (WB) or an indirect immunofluorescence assay (IFA). Over the past two decades, many advances in HIV diagnostics have been achieved: less complex rapid tests have been FDA-approved for screening at the point of contact (POC) with the patient, more sensitive and specific laboratory-based tests are available, and more complex rapid tests can detect HIV-2.

The “HIV Testing Algorithms: Status Report” describes a menu of HIV testing algorithms that have the potential to augment and provide alternatives to the algorithm currently used to diagnose HIV infection. APHL and CDC convened two working groups of HIV diagnostic subject matter experts to develop these new algorithms. This report describes algorithms to use in POC and laboratory settings, as well as supporting evidence, limitations, and additional data needed to substantiate the algorithms as of April 2009. This document does not contain recommendations from either CDC or APHL.

PROPOSED TESTING STRATEGIES FOR POC RAPID HIV TESTING

Most POC testing will be conducted with rapid HIV tests that are “waived” under the Clinical Laboratory Improvement Amendments of 1998 (CLIA).

- **Algorithm 1: Single Rapid Test (A1) for HIV Screening.**

This is the current algorithm, in which a single test is performed with an oral fluid or blood specimen. If the rapid test (A1) is reactive, a “preliminary positive” result for the presence of HIV-1 and/or HIV-2 antibodies is reported. A specimen should be collected for supplemental laboratory testing to confirm the results. If the initial test (A1) is non-reactive, a “negative” result for HIV-1 and/or HIV-2 antibodies is reported.

- **Algorithm 2: Two Rapid Tests (A1/A2) Performed in Sequence on Blood.**

Two different rapid test products are used sequentially, both on blood specimens, to improve the positive predictive value of POC testing. If rapid test A1 is non-reactive for HIV-1 and/or HIV-2 antibodies, a “negative” result for HIV-1 and/or HIV-2 antibodies is reported. If test A1 is reactive, a second test (A2) from a different manufacturer is performed. If both A1 and A2 are reactive, the result is reported as: “Presumptive positive for HIV-1 or HIV-2 antibodies; requires medical follow-up for further evaluation and testing.” If A1 is reactive and A2 is non-reactive, the result is reported as: “Inconclusive rapid test result; requires additional testing.”

- **Algorithm 3: Two Rapid HIV Tests (A1 oral/A2 blood) Performed in Sequence; If A2 Is Negative, A1 Is Repeated on Blood.**

In this algorithm, a reactive test performed on oral fluid (A1) is followed by a test from a different manufacturer (A2) performed on blood. If both A1 and A2 are reactive, the result is reported as: “Presumptive positive for HIV-1 or HIV-2 antibodies; requires medical follow-up for further evaluation and testing.” If the A2 test is nonreactive, the A1 test is repeated, but this time on a blood specimen. If the A1 test is reactive with both oral fluid and blood but the A2 test is non-reactive with blood, these results may represent false-positive A1 test results with two different specimen types or a false-negative A2 result. This should be reported as: “Inconclusive rapid test result; requires additional testing.” If the A1 test on the oral fluid specimen is reactive, but both blood tests (A2; follow-up A1) are non-reactive, then the result should be reported as: “Negative for HIV-1 and HIV-2 antibodies.” In this case the oral fluid test is considered a false-positive result and the blood tests are considered true-negative results.

- **Algorithm 4: Three Rapid HIV Tests (A1/A2/A3) Performed in Sequence on Blood. (A1, A2 and A3 must be different rapid tests.)**

The strategy of using three different blood tests in sequence has been proposed as a way to confirm results at the POC, but requires further validation before implementation. If A1 is reactive, another blood specimen should be obtained to perform test A2. If A2 is also reactive, the result is reported as: “Presumptive positive for HIV-1 and/or HIV-2 antibodies; requires medical follow-up for further evaluation and testing.” If the A2 test is negative, then another blood specimen should be tested using a different product, A3. If A3 is reactive, in conjunction with a reactive A1 result and a non-reactive A2 result, then the test results should be reported as: “Presumptive positive for HIV-1 and/or HIV-2 antibodies; requires medical follow-up for further evaluation and testing.” If A3 is negative, in conjunction with a reactive A1 result and a negative A2 result, the result should be reported as: “Inconclusive rapid test result; requires additional testing.”

PROPOSED TESTING STRATEGIES FOR LABORATORY HIV TESTING

In general, laboratories use assays categorized by CLIA as high complexity. However, some laboratories may choose to use rapid HIV tests (categorized under CLIA as moderately complex when used with serum or plasma) as one of the component assays of the laboratory testing algorithm.

- **Algorithm 1: HIV-1 Only Immunoassay, With Supplemental NAAT Option.**

This algorithm most closely reflects the current algorithm, a stand-alone HIV-1 immunoassay as the (A1) screening test followed by a (B1) supplemental test, either a Western blot (WB) or an indirect immunofluorescence assay (IFA). Samples that are repeatedly reactive using A1 are then tested with the B1 supplemental test; if B1 is also positive, the test results are reported as positive for HIV. Alternatively, after a repeatedly reactive A1 test, an individual nucleic acid amplification test (NAAT) (B2) can be used as a supplemental test for confirmation. NAAT (B2) can also be used to resolve negative or indeterminate WB or IFA (B1) results. If NAAT (B2) is negative but the sample was repeatedly reactive on the A1 test, B1 testing by WB or IFA is still required. If an acute HIV infection is suspected, refer to Algorithm 4 for guidance.

- **Algorithm 2: HIV-1/HIV-2 Immunoassay, With Supplemental NAAT Option.**

The inclusion of antigens designed to detect HIV-2 in FDA-approved diagnostic tests is increasingly common. Algorithm 2 offers the option of using B1 (HIV-1 WB or IFA) or B2 (HIV-1 NAAT) as a supplemental test on a specimen that is repeatedly reactive using A1. If B2 (NAAT) is not performed initially, it may be used to resolve indeterminate or negative B1 (WB or IFA) results. As in Algorithm 1, if B2 (NAAT) is negative but follows a repeatedly reactive A1, B1 testing (either a WB or IFA) is required. Algorithm 2 has the potential to detect more early-stage infections, but a negative NAAT result does not rule out infection: specimens that are reactive on an HIV-1/2 combo assay, but negative or indeterminate on supplemental testing, could represent HIV-2 infections. (See Algorithm 5 for more information). If an acute HIV infection is suspected, refer to Algorithm 4 for more information.

- **Algorithm 3: Dual HIV-1/HIV-2 Immunoassay.**

Dual immunoassays can potentially maximize the sensitivity of the algorithm to detect early and long-standing HIV infections while maintaining or improving specificity compared with the traditional EIA/WB algorithm. If both assays have similar sensitivity and specificity, the dual immunoassay could reduce the number of discordant results that occur with Algorithms 1 and 2. Algorithm sensitivity is dictated by the initial screening assay, thus the EIA or chemiluminescent immunoassay (CIA) with the best sensitivity should be used as A1. A negative A1 result is proposed to be reported as negative for HIV-1 and HIV-2 antibodies. (If acute HIV infection is suspected, Algorithm 3 should be followed with Algorithm 4.) The A2 test must be a different EIA or CIA than A1, with different antigen properties or binding/detection methods, to minimize concurrent non-specific reactivity in uninfected patients. If A1 and A2 are repeatedly reactive, the result is reported as: “Presumptive Positive for HIV-1 and/or HIV-2 antibodies and requires medical follow-up for further evaluation and testing.”

- **Algorithm 4: Acute HIV Infection Testing.**

Detecting HIV infection as early as possible after transmission can provide valuable information to patients and potentially prevent the spread of HIV. Testing of individual specimens by NAAT is useful for patients who have symptoms of acute retroviral infection and report recent high-risk exposure. The purpose of Algorithm 4 is to detect HIV-1 RNA in specimens with negative antibody results from an EIA, CIA or rapid test by either pooled or individual NAAT.

- **Algorithm 5: HIV-2 Testing.**

HIV-2 testing should be considered in cases where a client has potential epidemiologic risk factors for HIV-2 (i.e., sex partners from countries where HIV-2 is endemic, sex partners known to be infected with HIV-2, blood transfusion or non-sterile injection in countries where HIV-2 is endemic, needle sharing with a person from an HIV-2 endemic country or with a person known to be infected with HIV-2, children of women who have risk factors for HIV-2), clinical suspicion of AIDS in the absence of a positive test for antibodies to HIV-1; or an HIV-1 Western blot with unusual indeterminate patterns, such as Gag p55, p24 or p17 plus pol bands p66, p51 (RT) or p31/32 (integrase). At the time this status report was written, there was no FDA-approved supplemental test for HIV-2 confirmation.

There are many challenges associated with implementing the proposed algorithms, ranging from structural (policy, law) to operational (staff training, developing quality assurance protocols). The ultimate goal is to have an increased number of individuals tested accurately and, if infected, linked into medical care as soon as possible.

Key to Acronyms and Symbols

+ = Reactive; or, positive

- = Non-Reactive; or, negative

CI = confidence intervals

CIA= Chemiluminescent Immunoassay

EIA= Enzyme immunoassay

IFA=Indirect immunofluorescence assay

HIV-1/HIV-2 = HIV-1/2

NAAT= Nucleic Acid Amplification (HIV-1 RNA) Test

NPV = negative predictive value

POC = point of contact

PPV = positive predictive value

RR = repeatedly reactive

S/CO = signal-to-cutoff ratio

WB= Western blot