

Repeated False-Positive HIV Test Results in a Patient Taking HIV Pre-Exposure Prophylaxis

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Regular HIV testing is required to ensure the safety of HIV pre-exposure prophylaxis (PrEP). We describe and discuss a series of false-positive HIV test results from an individual receiving PrEP. The expansion of PrEP will likely result in greater numbers of false-positive test results that may pose challenges for interpretation.

Keywords. PrEP; pre-exposure prophylaxis; HIV testing; seroconversion; false-positive.

CASE REPORT

The patient was a 34-year-old African American cis-gender man who presented for regular interval testing for HIV and other sexually transmitted infections as follow-up for pre-exposure prophylaxis (PrEP) provided through the Public Health – Seattle & King County Sexually Transmitted Disease Clinic. He reported excellent adherence to tenofovir disoproxil fumarate co-formulated with emtricitabine (FTC/TDF) taken as PrEP for more than a year and had tested HIV-negative using the GS HIV Combo Ag/Ab EIA (Bio-Rad, Redmond, WA) 292, 194, and 98 days before the visit as part of regular follow-up with his provider when renewing his PrEP prescription. On the day of the visit, he enrolled in Project DETECT, an ongoing study recruiting persons seeking HIV testing to evaluate the performance of several HIV tests when performed at the point of care (POC) using unprocessed whole blood and oral fluid specimens [1, 2]. The patient was tested with 5 POC HIV rapid tests, a laboratory-based HIV antigen-antibody (Ag/Ab) screening test, and a quantitative HIV-1 viral load test, according to study protocol.

Results of the different HIV tests performed at POC and in the laboratory are shown in the Table 1. At the first study visit, the only abnormal test result was a positive HIV-1 p24 antigen result on the Determine HIV-1/2 Ag/Ab Combo (Alere, Inc., Boston, MA) test performed on a venipuncture whole-blood specimen. However, this result, in combination with negative POC antibody test results, was consistent with test results that could indicate acute HIV infection (AHI) [3], and he consented to participation in serial follow-up with repeat HIV testing at up to 9 visits over 70 days through Project DETECT, per protocol. He reported 5 male anal sex partners in the preceding 3 months, including 2 partners with whom he had condomless anal intercourse. The patient reported that 1 of these partners was HIV-negative; the other's HIV status was not reported. Although the patient denied any symptoms consistent with acute retroviral syndrome [4] and reported excellent adherence to PrEP (with only 2 missed doses reported in the past year), there was an initial concern that the positive POC test for the p24 antigen might represent true infection. These concerns subsided when both the GS HIV Combo Ag/Ab EIA and a quantitative RNA test (Abbott HIV-1 RNA, Abbott Molecular Diagnostics, Des Plaines, IL) from the initial visit were reported as negative a few days later.

Results of HIV tests performed over 70 days are shown in the Table 1. The patient continued to have positive Determine results, reacting only to the p24 antigen component of the test, throughout follow-up. On his fourth study visit (day 16 after initiation of Project DETECT), the patient tested HIV-2 indeterminate on the Geenius HIV 1/2 Supplemental Assay (BioRad Laboratories, Redmond, WA), performed on EDTA whole blood. This renewed concerns about true HIV seroconversion because the few cases of seroconversion among persons adherent to PrEP have shown anomalies in HIV testing, as reviewed in Smith et al. [5], and Geenius has been shown to produce HIV-2-indeterminate results in patients with acute HIV-1 infection [6]. To further attempt to resolve the patient's HIV status, at day 21 a specimen was submitted for a different antigen/antibody test ARCHITECT HIV Ag/Ab Combo (Abbott Laboratories, Chicago, IL), and at day 29 a total nucleic acid test (Cobas Taqman HIV-1, Roche Molecular Diagnostics, Pleasanton, CA) was performed on a whole-blood specimen. Both of these additional tests were also nonreactive. The patient remained on PrEP through completion of 70 days of follow-up, and the study ultimately considered him HIV-negative but with unresolved reactivity to the p24 antigen component of the Determine test.

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Table 1. Serial Follow-up of a Patient With Difficult-to-Interpret HIV Test Results who Did Not Acquire HIV Infection While Taking TDF/FTC for PrEP

Patient	Days From First Positive Test	Determine HIV-1/2 Ag/Ab Combo ^a	Instrumented Ag/Ab Tests ^{b,c}	HIV-1 Ab-Only Point-of-Care Tests ^d	Genieus HIV-1/2 Supplemental Assay ^e	HIV-1 Viral Load, Log ₁₀ Copies/mL ^{f,g}
34yo MSM; Seattle	0	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive ^e	TND ^f
	8	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive ^e	TND ^f
	12	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive ^e	TND ^f
	16	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive, ^e HIV-2 indeterminate ^e	TND ^f
	21	Ab nonreactive, Ag reactive	Nonreactive ^{a,b}	All nonreactive	Nonreactive ^e	TND ^f
	29	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive, ^e HIV-2 indeterminate ^e	TND ^{f,g}
	36	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive, ^e HIV-2 indeterminate ^e	TND ^f
	52	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive, ^e HIV-2 indeterminate ^e	TND ^f
	57	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive ^e	TND ^f
	70	Ab nonreactive, Ag reactive	Nonreactive ^a	All nonreactive	Nonreactive ^e	TND ^f

More information on these tests, including links to current FDA-approved package inserts, is available at <https://www.cdc.gov/hiv/testing/laboratorytests.html>.

Abbreviations: Ab, HIV-1/HIV-2 antibody; Ag, HIV-1 antigen; MSM, gay, bisexual, or other man who has sex with other men; ND, not done (ie, test not performed); PrEP, HIV pre-exposure prophylaxis; TND, target not detected (ie, test was negative for HIV-1).

^aDetermine HIV-1/2 Ag/Ab Combo, Alere, Inc. (package insert revision 11/2016).

^bBioRad GS HIV Combo Ag/Ab, BioRad Laboratories, Redmond, WA (package insert revised 7/2011).

^cARCHITECT HIV Ag/Ab Combo, Abbott Diagnostics, Chicago, IL (package insert revised 12/2009).

^dOraQuick ADVANCE Rapid HIV-1/2 Antibody Test, OraSure Technologies, Inc. Bethlehem, PA (package insert revised 03/2016B), BioLytical Insti HIV-1 rapid test Richmond, BC, Canada (package insert revised 2017), Chembio DPP HIV-1/HIV-2 rapid test, Chembio Diagnostics, Medford, NY (package insert revised 9/2016).

^eGenieus HIV 1/2 Supplemental Assay, BioRad Laboratories, Redmond, WA: a) anticoagulated (EDTA) whole-blood specimen; b) fingerstick whole-blood specimen (package insert revised 01/2013).

^fAbbott real-time HIV-1 RNA quantitative viral load assay, Abbott Molecular Diagnostics, Des Plaines, IA (package insert revised 12/2011).

^gCobas Taqman HIV-1 test v2.0, total nucleic acid (performed on whole blood), Roche Molecular Diagnostics, Pleasanton, CA (laboratory developed, research-use-only test validated by the University of Washington HIV laboratory).

DISCUSSION

This case provides an example of how the interpretation of abnormal and ambiguous HIV screening test results can become more challenging in the context of PrEP. Initially, the patient tested reactive only for the p24 antigen on the Determine test and negative on all other POC antibody tests. In the context of a person at high risk for acquiring HIV infection, this result could indicate a recent infection, as the “window period” for antigen detection is shorter compared with antibody [3]. There has been much recent discussion about the potential for false-negative test results for persons taking PrEP [5]. However, in the context of PrEP, excellent reported adherence, and regular follow-up for interval HIV and sexually transmitted infection testing, HIV acquisition would be unlikely [7–9]. False-positive results occur with all HIV screening tests and should be expected. Furthermore, the Determine test has been observed to have lower specificity than other POC devices, and thus more false-positive test results may be seen with this test [10]. It is important that all positive screening test results be confirmed by using additional tests according to established test algorithms [5].

False-positive results are uncommon, and although repeat false-reactivity on the same test over several months has been reported anecdotally, it has not been fully documented in the literature. For example, in the iPrEx PrEP trial, there were 8 reactive test results that were determined to be false-positive among 50 260 tests of 2499 study participants [11]. These 8 reactive tests occurred among a total of 4 participants; all occurred among patients in the control arm, 1 of whom was repeatedly false-reactive on the Determine antibody-only test (a different test manufactured by Alere, Inc., not available in the United States) 4 times before leaving the study (R. Grant, MD, MPH, unpublished data). Likewise, in the Partners PrEP study, 23 participants were reported to have had false-positive test results at more than 1 study visit [9]. Although the POC tests used in that study were reported to detect only HIV antibody [9], the exact tests used and the timing, sequence, and resolution of false-positive results have not yet been described. Although both POC and laboratory-based tests that detect the HIV-1 p24 antigen may have lower specificity than antibody-only assays [10], the POC test presents an additional complication because providers

and patients receive a preliminary result before laboratory testing can confirm that result [12].

In this case, the combination of negative laboratory-based tests for p24 antigen, HIV antibody, and HIV RNA from venipuncture specimens support the working hypothesis that the persistent reactivity to the p24 antigen on the POC test was most likely a false-positive result. However, given the patient's exposure to antiretroviral medications, it was important to interpret test results with caution. At least 4 breakthrough PrEP infections with ambiguous HIV test results have been reported among persons using oral daily FTC/TDF or TDF. These cases required multiple follow-up tests to resolve their ambiguous test results [5]. Of these 4 individuals, 1 had a reactive Ag/Ab EIA but a negative antibody-only supplemental test and a quantitative HIV RNA with signal detected but below 20 copies/mL. Another had a reactive Ag/Ab screening test but a negative Western blot, no signal detected on a quantitative HIV RNA, and a follow-up test for HIV DNA that was also negative. Although these results were nearly identical to those of the present case, we elected to continue PrEP while pursuing additional testing, and negative Ag/Ab test results from different manufacturers, as well as a negative total nucleic acid tests that can detect integrated proviral DNA [13], provided additional reassurance that the Determine test was false-positive. At present, only lab-developed tests are available for total HIV-1 nucleic acid and proviral DNA testing, as well as assays that directly detect biomarkers of PrEP adherence [14, 15] and these tests are only available at select laboratories. Biomarker assays that detect TDF or FTC directly might prove helpful for patients where adherence to PrEP is not as well documented as in this case. Further, in a situation where PrEP adherence is suspected or known to be poor, or where there is high likelihood of recent infection, alternatives such as stopping PrEP to assess for viral rebound or initiating a fully suppressive regimen, as would be prescribed for postexposure prophylaxis or treatment initiation, may also be considered [5]. When additional testing is available, it is our opinion that any concern for HIV infection in someone on PrEP, particularly someone with recent exposures and symptoms of AHI, warrants additional evaluation that might include use of a total nucleic acid test, an antigen-antibody test with a different target (or at least from a different manufacturer if the target of a particular test is unknown), and consultation with experts in HIV testing of PrEP patients. Whether or not additional diagnostics are available, the decision to continue PrEP, add a third antiretroviral agent to create a suppressive regimen, or discontinue all medications (reviewed by Smith et al. [5]) should be based on test results and the likelihood of true HIV infection according to reported or measured PrEP adherence, exposure history, and the presence or absence of clinical symptoms of AHI.

An expected consequence of quarterly HIV testing among the increasing numbers of persons on PrEP [16] will be the

increasing numbers of false-positive HIV test results that require extra evaluation and investigation. As a hypothetical example, in a PrEP clinic with 250 patients testing quarterly, assuming that all remain HIV uninfected, and the use of an HIV test with 99.5% specificity, there will be 5 false-positive tests per year. In the Partners PrEP Study, 110 (69.2%) of 159 reactive POC tests among persons randomized to PrEP were false-positive test results [9], emphasizing that in a population taking PrEP with high adherence, false-positive results are likely to outnumber true-positive results. Out of the thousands of people taking PrEP for HIV prevention, there have been no more than 4 people with documented HIV acquisition despite PrEP adherence commensurate with >90% protection against HIV [5, 8]. Most people prescribed PrEP who acquire HIV infection will do so due to medication nonadherence [7–9], and the vast majority of anomalous HIV test results in persons adherent to PrEP will be false-positive. Though that knowledge should be reassuring, clinicians are still obligated to rule out HIV infection in every case, and they are likely to evaluate many cases similar to the one we describe here.

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