Instructions for Specimen Collection in Cases Involving Special Supplemental Testing at the Public Health Laboratory:

Supplemental Testing to Resolve Equivocal Diagnostic Test Results
Update July 2010

Background—Diagnosis of HIV

In the overwhelming majority of cases, HIV testing using commercially available 3rd generation EIA with supplemental WB clearly establishes or rules out a diagnosis of HIV infection. False negative tests generally occur in the pre-antibody "window period" and can be resolved by repeat testing of a new specimen drawn at a later date. False positive tests that occur during pregnancy, intercurrent infection or inflammation, autoimmune disease, or for other reasons, are also generally resolved by repeat testing.

Occasionally, however, a patient will have a series of indeterminate tests and/or a series of tests with contradictory results, e.g., one or more positive tests followed by negative test(s). In other cases a patient with deteriorating immune function and/or opportunistic infection tests repeatedly negative for HIV-1. Particularly among patients from West African countries, where HIV-2 is endemic, it is important to rule out HIV-2. Although most commercial laboratories and some hospitals now routinely screen for HIV-2 using an HIV-1-2+O EIA test, specific testing for diagnosis and/or confirmation of HIV-2 is not available in most settings, nor is testing to establish a diagnosis of a Group O or N infection. Establishing a definitive diagnosis is vital to the health and well-being of the patient as well as for prevention of secondary transmission and public health reporting.

Availability of Additional Laboratory Testing

The NYC Department of Health and Mental Hygiene offers laboratory services to assist NYC physicians to establish or rule out a diagnosis of HIV in these cases and any other cases where providers have questions about diagnostic test results. A standard testing algorithm is followed. In the order presented, the following tests are conducted: (1) HIV 1-2+0 EIA, (2) HIV-1 specific EIA, (3) HIV-2 specific EIA, (4) HIV-1-2 multispot, (5) HIV-1 WB, (6) HIV-2 WB, (7) HIV-1 quantitative RNA PCR, (8) HIV-2 DNA PCR and, if indicated, (9) CD4 flow cytometry.

Should viral culture or genetic sequencing be needed to establish the diagnosis we can also make available laboratory services provided by CDC. All of these services are offered free of charge to NYC providers. Assistance with resolution of equivocal HIV-1 test results and specific diagnosis of HIV-2 is a joint effort of the Virology and Immunology Division of the Public Health Laboratory (PHL) and the HIV Epidemiology and Field Services Program.

Whom to Contact

Please contact one of the two contacts listed below to discuss the case and make arrangements for specimen collection and pickup:

Dr. Lucia Torian (*Deputy Director, HIV Epidemiology and Field Services Program*) at 212-442-3461. Email address: ltorian@health.nyc.gov

or

Mr. Robert Pirillo (*Community Coordinator, Division of Virology and Immunology, PHL*) at 212-447-2881. Email address: rpirillo@health.nyc.gov.

Dr. Torian and Mr. Pirillo work closely together and are in constant communication. A call to either one will result in the other being notified of the case immediately.

Back-up Personnel to Contact

In the event that you cannot reach Mr. Pirillo or Dr. Torian, call:

At the HIV Epidemiology Program: Lisa Forgione, 442-3453 or lforgion@health.nyc.gov.

At the PHL: David Yang, 212-447-2864 or dyang@health.nyc.gov.

Procedure for Specimen Collection

(1) Call either Dr. Torian or Mr. Pirillo to confirm the date and time of the appointment at which you plan to draw specimens for HIV testing. Blood should be drawn as early as possible during the day.

(2) Complete the NYCDOH "Request for HIV Analysis" specimen submission form. Most facilities have these forms on hand. If you do not have one, we will bring one with us for you to fill out. If we are sending you this instruction protocol via email, we will also attach the PHL laboratory requisition. If you are requesting differential diagnosis of HIV-2, we will also send our HIV-2 health advisory. Finally, if we are recommending that you consider genetic sequencing or other testing performed at CDC, we will also ask you to fill out a CDC "DASH" form and will provide it by email.

(3) Draw the following tubes of blood:

(a) One (1) red top (serum) tube (no anticoagulant)
(b) Two (2) yellow top tubes with ACD (*do not use heparin!*)
(c) Two (2) lavender top tubes with ACD or EDTA (*do not use heparin*)

(4) Draw full tubes.

(5) Label the blood with the patient's full first name, full last name, date of birth, sex, and the specimen draw date. Place a label on each tube.

(6) Rack and *maintain all tubes at room temperature*. Do not refrigerate. Keep undisturbed to the extent possible.

(7) The specimens and submission forms will be picked up by our laboratory staff at the time of the appointment. In most cases pickup will be made by the manager of the HIV Epidemiology Laboratory; in some cases a laboratory messenger will be sent. We must pick up the blood when it is as fresh as possible, i.e., within 6 hours of collection, and it must be transported back so that it arrives at the PHL no later than 2:00 p.m. on the draw date.

Laboratory Protocol

The PHL will conduct its full "special study" protocol as described above. If necessary, we will consult with CDC on interpretation of test results and recommendations for future testing. Based on the test results, we will consult with you, the attending physician, as to whether you would like further testing performed by CDC.

Results

Testing generally begins on the date of the blood draw, depending upon how early the specimens are brought back to the PHL. The full PHL algorithm generally takes two weeks to complete. Supplemental testing by CDC (if needed) requires additional time. Dr. Torian or Mr. Pirillo will notify the attending physician by telephone that the results are available. At the time of notification they will also make arrangements to provide written results by a secure means. Results (identified by patient ID# to protect confidentiality) can be FAXed, but we request that you (1) send a written request to 212-447-2877, (2) stand by to receive the FAX personally, and (3) FAX back a cover sheet acknowledging receipt of the report.

Additional Testing by CDC

In the event that we recommend additional testing and/or review of local results by CDC, and the attending physician concurs with the recommendation, we will contact CDC and communicate their recommendations, testing plans, and results as they become available.

Questions?

If you have any questions, or if you have a particular case that you would like to discuss, please do not hesitate to contact us:

Dr. Lucia Torian (*Deputy Director, HIV Epidemiology and Field Services Program*) at 212-442-3461. Email address: ltorian@health.nyc.gov

or

Mr. Robert Pirillo (*Community Coordinator, Division of Virology and Immunology, PHL*) at 212-447-2881. Email address: rpirillo@health.nyc.gov.

You may also contact:

Dr. Amado Punsalang (*Director, HIV Testing Laboratory, Division of Virology and Immunology, PHL*) at 212-447-2864. Email address: apunsala@health.nyc.gov.