Saint Luke's Regional Laboratories
Clinical Laboratory Letter
October 2013

HIV Western Blot Obsolete

For the past 25 years, the traditional HIV testing algorithm has consisted of a two step strategy including HIV-antibody screening with Western blot confirmation of reactive results. However, improvements in sensitivity of newer generation HIV-antibody screens have created the dilemma of falsely-negative Western blot assays. Third generation HIV-antibody immunoassays can detect seroconversion as early as 22 days after infection, while Western blot may not show reactivity until 4 weeks or more.

Therefore, CDC and Clinical Laboratory Standards Institute (CLSI) have designed a new HIV testing algorithm that eliminates Western blot confirmation. The first test in the new algorithm remains the combination HIV-1/HIV-2 antibody immunoassay. The follow-up confirmation test for reactive results is a new HIV-1/HIV-2 differentiation immunoassay, known as Multispot, which detects seroconversion earlier than Western blot and eliminates most indeterminate results that occur due to nonspecific reactivity from alloantibodies.

Effective immediately, all specimens that are reactive by initial HIV-1/HIV-2 antibody immunoassay will be forwarded to a reference laboratory for confirmation by Multispot testing. Specimens that do not confirm positive by Multispot may require further analysis by HIV RNA PCR.

Autoantibodies for Thyrotoxicosis

All forms of autoimmune thyrotoxicosis are caused by the production of autoantbodies to the thyrotropin stimulating hormone receptor (TSHR-Ab), which are also known as long-acting-thyroid-stimulator (LATS) or thyroid-stimulating immunoglobulins (TSI). These autoantibodies bind to the thyrotropin receptor leading to thyroid stimulation independent of TSH.

Physicians commonly order both TSHR-Ab and TSI, which is not necessary, because TSHR-Ab test detects both TSI and TSHR-Ab. TSHR-Ab test has a shorter turnaround time, is less expensive, and has excellent correlation with the TSI assay. Sensitivity is 97% and specificity is 99% for detection of Graves disease.

Free Light Chain Confusion

Serum free light chain analysis is the newest test added to the workup of plasma cell disorders. This analysis includes quantitation of both the kappa and lambda free light chains, as well as calculation of the kappa to lambda ratio. Our laboratory has received several inquiries regarding the interpretation of serum free light chain results following the recent change in reference laboratories. Previously, serum free kappa and lambda light chain results were reported in units of mg/dL. Now they are reported in units of mg/L. Because a liter (1000 mL) is ten times more than a deciliter (100 mL), results are 10 times higher than previously. Old results can be compared to new results by multiplying the old results by a factor of 10. The kappa to lambda ratio is not affected.

Optimized Trichomonas Testing: Male Specimens Too!

Trichomoniasis is a sexually transmitted infection caused by the parasitic protozoan *Trichomonas vaginalis*. According to the latest CDC data, infections with *T. vaginalis* have exceeded the prevalence of *Chlamydia*, especially in the 25+ age group. Complications of untreated *Trichomonas* infections include pelvic inflammatory disease, pregnancy difficulties including premature labor, and infertility. Unfortunately, up to 70% of *Trichomonas* infections are asymptomatic, in both sexes. Symptoms of infection in women include vaginitis, urethritis, or cervicitis. Male symptoms include urethritis or prostatitis.

Several options with variable performance are available for diagnosis of trichomoniasis in women. Wet prep and culture have the lowest sensitivity (35%-80%) and require immediate specimen transport for optimal results. Molecular diagnosis, such as the DNA probe hybridization test offered by Saint Luke's Microbiology (BD Affirm VP), improves sensitivity to 92% and specificity to 99%. Affirm

vaginitis testing requires a vaginal swab specimen, submitted in Affirm Ambient Temperature Transport System, supplied by SLRL.

Diagnosis of trichomoniasis in males has been problematic, as neither wet prep nor Affirm can be performed on male specimens. Recently, PCR testing has become available for male urine specimens. For optimal results, the patient should not have urinated for at least one hour prior to specimen collection. A first-catch urine specimen (20-30 mL of the initial urine stream) should be submitted in a preservative-free sterile urine collection cup. Testing is performed by a reference laboratory.

New RBC Antibody Titer Method

The Blood Bank at Saint Luke's Hospital performs red blood cell antibody titers for the entire health system. On November 4th 2013, the method for determining antibody titers will change from a traditional tube to an automated gel method, which decreases analytical time by 45 minutes and is more sensitive.

Because of increased sensitivity, physicians will notice an increase in antibody titer for clinically significant antibodies. If a patient is currently being followed and has had titers reported before November 4, the next titer will be performed by both the old and new methods and both results will be reported for comparison. Thereafter, only the titer of the new method will be reported.

New Rapid Flu Antigen Test

A new rapid influenza antigen test, the BD Veritor System, will be utilized throughout Saint Luke's Health System laboratories this season, for identification and differentiation of influenza A & B. Compared to previous assays, this test has enhanced sensitivity and specificity partially because results are interpreted by an optical reader, which improves objectivity.

Sensitivity is lower than specificity for all rapid influenza antigen tests; therefore false negative results are more likely than false positive results. False-positive results are most likely early in the influenza season, when prevalence in the community is low. Influenza specimens tested within 3-4 days of illness onset are more likely to yield true positive results. Suspected false negative results should be followed by PCR testing, when illness is severe or when otherwise clinically indicated.

Nasopharyngeal swabs for rapid influenza testing should be collected on FLOQSwabs (flocked swabs) and submitted in M6 viral transport media. Dry swabs cannot be tested. Nasal washes and aspirates are also acceptable for testing, but bronchoscopy specimens cannot be tested. Specimens should be transported to the laboratory as soon as possible, but can be refrigerated for up to 72 hours prior to testing, if necessary.

New Swabs for Culture Collection

Saint Luke's Microbiology is working to simplify specimen collection through elimination of multiple swabs & transport devices. A new flocked swab for bacterial specimens, called the Copan eSwab Transport System, has been chosen for use throughout Saint Luke's Health System. All bacterial cultures, both aerobic and anaerobic, can be collected using the Copan eSwab Transport System. This system can also be used for wet mounts, group B strep PCR, and fungal cultures.

Please note that all swab-collected viral cultures, rapid influenza tests, rapid RSV tests, and respiratory PCR tests still require M6 transport media & should continue to be collected with the Copan flocked swab designated FLOQSwab. Swab specimens are inadequate for AFB culture and will be rejected for testing with the exception of pediatric specimens.

Flocked swabs have enhanced surface area and have been shown to be superior to wrapped swabs for pathogen recovery in multiple settings. The flocked swab type is recommended by the American Society for Microbiology and Clinical Laboratory Standards Institute.

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