



SEPT/OCT 2008

PATHOLOGY & LABORATORY MEDICINE NEWSLETTER

NEWS YOU CAN USE

New Tests Implemented in Special Chemistry Section

Blood Lead was brought in-house in September. Anti-CCP antibody will be brought in-house in October, paired with a new ELISA method for Rheumatoid Factor (see separate articles in this issue).

Stanford Clinical Lab May Still Serve Community Physicians

The transfer of Stanford Clinical Lab's outreach business to LabCorp in August created some confusion for community physicians. Patients from the community who are being admitted to either LPCH or Stanford Hospital (or referred to a hospital clinic) in the near future may utilize the services of our laboratory. These patients may visit Patient Service Centers (PSCs) at Stanford Hospital; Blake Wilbur Building; Menlo Clinic; 730 Welch Road (for pediatric patients); and 770 Welch Road (for obstetric patients). Stanford Clinical Lab no longer operates additional PSCs.

Testing ordered in a community physician's office that is required "stat" may also be sent to Stanford Clinical Lab, as well as surgical pathology, cytopathology, hematopathology and some esoteric testing. However, specimens for basic laboratory testing collected in a community physician's office from patients who do not have an active registration at either hospital must

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GETTING THE LEAD "IN": Blood Lead Test Now In-House Using ICP-MS

Raffick Bowen PhD – Associate Medical Director of Clinical Chemistry & Immunology

Lead is no longer the environmental threat it once was, thanks to federal regulations removing it from paint, gasoline and metal used in food packaging. These efforts are reflected in the action thresholds for blood lead levels recommended by the Center for Disease Control (CDC): lowered from 60 µg/dl in the 1960s to the 10 µg/dl target used today. Nonetheless, even without contaminated imports from other countries, lead may pose a risk. In addition, much lead remains in the environment years after its initial use.

Although adults may be at risk, children have always been a primary focus of screening for lead exposure. Acute lead poisoning in children typically presents with severe abdominal pain, vomiting and diarrhea as well as neurological symptoms such as seizures or coma. In chronic lead poisoning, signs and symptoms include anemia (with basophilic stippling, see Figure 1), developmental delay, hyperactivity, and other behavioral disturbances. Major risk factors include low socioeconomic status and foreign adoption.

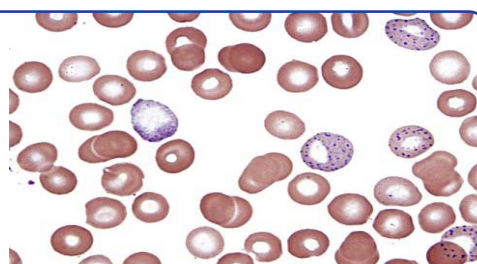


Figure 1:

Lead interferes with heme synthesis at several points resulting in a microcytic, hypochromic anemia similar to that seen in iron deficiency. In lead poisoning, however, one may also see characteristic coarse basophilic stippling, due to aggregated ribosomes.

CMS mandates that all children enrolled in Medicaid receive blood lead screening. Under California law, children with identified risk factors must be screened twice (at age one and again at age two years).

Stanford's Clinical Laboratory stopped performing the **test for blood lead** (using anodic stripping voltammetry) in 2005 because of a new requirement that all blood lead testing be reported to the state of California's Department of Health Services electronically, which our information system could not do at the time. For the past few years, we have been referring specimens to an outside reference laboratory that used a method called inductively-coupled plasma mass spectrometry (ICP-MS, see Figure 2). This year, we implemented our own ICP-MS instrument and, beginning in September, brought this test in-house (**order code: LEADBL**).

We continue to use the CDC's action threshold as our reference range and will call all results greater than 10 µg/dl to the ordering physician. Results greater than 45 µg/dl require immediate re-testing and consideration of chelation therapy.

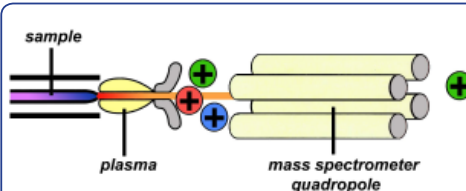


Figure 2:

In the ICP-MS lead method, diluted whole blood is pumped into a "plasma" (ionized gas) and atomized. The atoms absorb energy, release an electron and become positively charged. Using mass-to-charge ratio, the mass spectrometer allows only lead atoms through to the detector.

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NEWS YOU CAN USE (CONT.)

be considered outreach business and cannot be accepted for testing at Stanford Clinical Laboratory. These samples should be sent to LabCorp (or another reference laboratory of choice).

Capillary Blood Glucose Improvements

Our Point-of-Care-Testing section replaced the instruments for capillary blood glucose at both hospitals in July. Advantages of the new "Precision Xceed Pro" (PXP) include more reliable scanning of patient barcodes; new moisture guards to prevent blood contamination of the instrument; and an enhanced software system for keeping track of patient results.

Laboratory Information System Upgraded

On the heels of the EPIC[®] Hospital Information System implementation, the Stanford Clinical Laboratory's Sunquest[®] Laboratory Information System underwent a major upgrade in August. This will allow us to have significantly greater storage capacity. In addition, the Microbiology and Virology sections may now utilize a "paperless" GUI (graphical user interface); all patient-related inquiry functions are centralized; the specimen labels have much more information embedded in the barcode; and several enhancements to "call back" (an automated process for notifying physicians and nurses about important test results) will be possible in the near future.

LABletter

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BETTER DIAGNOSIS OF RHEUMATOID ARTHRITIS:

New Method for Rheumatoid Factor and Addition of Anti-CCP

Jim Faix MD – Medical Director of Clinical Chemistry & Immunology

Despite its name, rheumatoid factor (RF) is not a specific laboratory test to diagnose rheumatoid arthritis, one of the most prevalent autoimmune disorders. An antibody against itself (reacting with autologous immunoglobulin), RF is characteristic of rheumatoid arthritis. However, it is neither specific (it may be increased in many chronic infectious and inflammatory disorders) nor sensitive (it may be absent in up to 20% of rheumatoid arthritis patients).

Just as the past decade has seen advances in the treatment of rheumatoid arthritis, new approaches to the laboratory diagnosis have evolved. One of the key new markers is **antibody to cyclic citrullinated peptide** or anti-CCP (**order code:CCP**). Anti-CCP appears to be very specific for rheumatoid arthritis. It may also be positive earlier in the course of the disease (allowing for earlier intervention). Beginning this month, we are bringing the test for anti-CCP in-house and are pairing it with a **new method for Rheumatoid factor (order code: RHF)**.

The ELISA for anti-CCP antibody detects the presence of a heterogeneous group of antibodies (originally detected by immunofluorescence) directed against connective tissue proteins which have undergone an enzymatic post-translational modification turning arginine residues into citrulline. The "cyclic" part of the name refers to the fact that the synthetic peptides used in the ELISA test are formed into a loop to better allow antibody recognition (see figure 3).

We are using the original anti-CCP assay, with cyclic peptides resembling citrullinated filaggrin

(a large protein that associates with keratin filaments and may cause maturing keratinocytes in the skin to undergo apoptosis as the stratum corneum develops.) Across multiple studies, this assay has exhibited a sensitivity of approximately 70% and a specificity of approximately 95% for rheumatoid arthritis. Recently, some companies have created a new test by adding several additional citrullinated peptides, in an attempt to enhance the test's sensitivity. It is clear that a variety of citrullinated proteins may be autoantigens in rheumatoid arthritis. But reports which have looked at the newer assays have found that the slight increase in sensitivity may be offset by a decrease in specificity.

Although a recent meta-analysis (see Ann Int Med 2007; 146:797-808) concluded that anti-CCP antibody alone probably performed as well as both tests combined, RF remains a component of the American College of Rheumatology criteria for diagnosis of rheumatoid arthritis. Previously, we have used a turbidimetric assay for RF; in conjunction with bringing anti-CCP in-house, we are moving to ELISA in order to enhance the test's sensitivity.

Both RF and anti-CCP will continue to be orderable individually, but, beginning this month, there will also be a new battery including both tests. This **new order code is RAAS, for Rheumatoid Arthritis Antibody Screen**. Certainly, measuring both antibodies optimizes sensitivity (especially for early diagnosis) and, if both are positive, the diagnosis is strongly suggested.

Figure 3:

By creating a loop in a synthetic peptide that contained the key epitope recognized by anti-CCP antibodies (an arginine amino acid residue turned into citrulline), the sensitivity of the ELISA assay was increased. These autoantibodies are much more specific than rheumatoid factor for rheumatoid arthritis and may even be detectable before the onset of clinical symptoms.

