REX HEALTHCARE

Laboratory Bulletin...

Updates and Information from Rex Healthcare and Rex Outreach

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Ionized calcium, specimen change for outpatients

Effective immediately, the specimen requirement for outpatient ionized calcium is changed to serum. The preferred procedure is to draw a "gold top" serum separator tube (red top tube is also acceptable), allow the tube to clot 15 to 30 minutes at room temperature, centrifuge and refrigerate until courier pick-up. Uncentrifuged specimens are acceptable, if refrigerated. ***It is very important not to open the tube or expose the serum to air. If possible draw a separate tube for ionized calcium when other serum tests are being ordered.*** When kept refrigerated (and unopened) at 4°C, serum specimens are stable at least 48 hours.

This change is based on recommendations made by the National Center for Clinical Laboratory Standards (NCCLS) and stability experiments performed at Rex Laboratory. It will provide increased ease of specimen handling as well as improved specimen integrity.

Please make the following changes in the Rex Ancillary Services Handbook (1994), page 165; **Calcium, Ionized, Blood,** *Specimen* change to **serum;** *Container* change to **gold top (serum separator) or red top tube**. Causes for rejection: delete **clotted**.

The above comments pertain to Outpatient specimens. For Inpatients, either heparinized blood gas syringes or green top (heparinized) Vacutainer® tubes are recommended. Deliver to Laboratory immediately.

Robert B. Brainard, Ph.D.

Susceptibility testing for *Haemophilus*

H. Influenzae causes a variety of human diseases, including epiglottitis, sinusitis, otitis media, meningitis, septicemia, cellulitis and pneumonia. Until 1974, *H. Influenzae* was considered to be susceptible to ampicillin.[1] Beta-lactamase producing *H. influenzae* which are resistant to ampicillin are now common. For this reason, the laboratory performs a beta-lactamase test on all isolates.

H. influenzae is predictably susceptible to the third generation cephalosporins. For this reason, routine susceptibility testing of *Haemophilus* will be limited to the beta-lactamase test.

Karl T. Kleeman, Ph.D.

[1] Cumitech 6A, American Society for Microbiology, February, 1991.

International normalized ratio and protimes

The international normalized ratio (INR) is an attempt to enable patients who are on Coumadin therapy to have prothrombin times run at different labs and still have comparable results. This is more important today because patients' insurance companies will not pay for the test unless sent to their chosen lab. Most professional organizations have recommended using the INR rather than the PT ratio to monitor patients on Coumadin therapy. The INR is equal to patient PT divided by the lab mean value raised to the power of the International Sensitivity Index (ISI) of the thromboplastin used in the prothrombin time.

The National Committee for Clinical Laboratory Standards has recommended that laboratories use a thromboplastin reagent with an ISI value that approaches 1.0. Since most older reagents have ISI values closer to 2.0, the newer recommended reagents are more sensitive to Coumadin. The increased sensitivity results in a longer PT with the same dose of Coumadin and provides a wider therapeutic range. The INR compensates for this increased sensitivity when comparing prothrombin times run in different labs or in the same lab when reagents are changed. It is superior to monitoring a patient using the PT ratio.

The recommended therapeutic level of anticoagulation for most patients on Coumadin is an INR of 2.0 to 3.0. For patients with mechanical heart values the target INR range is 2.5 to 3.5. As shown in the table below, as the ISI decreases (increased sensitivity) the INR decreases. Note the PT ratio stays the same even though there is a change in the level of anticoagulation as reflected by the INR. It is recommended that the INR be used instead of the ratio when monitoring patients on Coumadin therapy.

INR	RATIO	ISI	SEC	MEAN
1.4	1.2	2.1	14	12
1.3	1.2	1.9	14	12
1.3	1.2	1.5	14	12
1.8	1.3	2.1	16	12
1.7	1.3	1.9	16	12
1.5	1.3	1.5	16	12
2.3	1.5	2.1	18	12
2.2	1.5	1.9	18	12
1.8	1.5	1.5	18	12
2.9	1.7	2.1	20	12
2.6	1.7	1.9	20	12
2.2	1.7	1.5	20	12

The routine use of the INR when monitoring Coumadin is important since the Rex Laboratory will change in approximately one month to a new instrument and a more sensitive reagent for prothrombin times. This will result in a different therapeutic range when the PT result is expressed in seconds. The therapeutic range when expressed as the INR will remain the same, i.e. 2.0 to 3.0.

Stephen Chiavetta, M.D.

Blood culture utilization

To assure adequate volume, initially two blood cultures should be ordered on adults. One blood culture is generally sufficient for newborns and babies. The following table was recently published.[1]

CONDITIONS AND PROTOCOLS FOR COLLECTING BLOOD SPECIMENS

Clinical Condition	Protocol	Comment
Severe septicemia, meningitis, osteo-myelitis, arthritis, pneumonia	Two cultures before therapy	One 10- to 20- ml sample from each arm
Subacute bacterial endocarditis	Three cultures in 24 hours	Collect two at start of fever spikes. Collect three more if the first three are negative after 24 hours.
Acute bacterial endocarditis	Three cultures within 1-2 hours before therapy	
Low-grade intravascular infection	Three cultures in 24 hours	Collect two at first sign of febrile episode.
Febrile episodes	No more than three cultures	Bacteremia may precede fever by 1 hour

1. Collect blood before the administration of antibiotics.
2. Ideally, collect blood in the hour before a predicted fever spike or as the spike begins.
3. A total of three cultures per 24 hours is usually sufficient to rule out bacteremia or endocarditis.
4. Blood for culture should not be withdrawn through an indwelling intravenous or intra-arterial catheter unless it cannot be obtained by venipuncture, or unless specifically ordered by the requesting physician.
5. If possible, draw blood below an existing intravenous line to prevent dilution of the blood with infusion fluid.

If a blood culture becomes positive, two "test of cure" cultures may be ordered 48 hours after the last positive blood culture. Two additional bloods may be drawn every 48 hours until negative.[2]

Karl T. Kleeman, Ph.D.

[1] *A Guide to Specimen Management in Clinical Microbiology,* J. Michael Miller, ASM Press, 1996.
[2] Adapted from UNC recommendations.

Use of the ASO and anti DNase B tests

The ASO (antistreptolysin O) and anti DNase B tests may be used for the confirmation of poststreptococcal sequelae, rheumatic fever and glomerulonephritis. An elevated ASO titer is reliable in patients with rheumatic fever (85% of patients) while the anti DNase B test is needed for diagnosing cases of glomerulonephritis.[1] Together, these two tests represent the best available serologic screening for poststreptococcal sequelae.

If a serum sample is obtained within 2 months of onset, approximately 80% of patients with acute rheumatic fever will have an ASO titer of greater than 200. By using a second test such as the anti DNase B test, the percent showing a serological response increases to 90%. [2]

The Rex Laboratory normal range for ASO is 0-250 IU. This is based on the reagent manufacturer's recommendations and verified by on site testing.

Although the ASO may be elevated, the diagnosis of acute glomerulonephritis is best supported by elevated antibody to anti DNase B. Normal ranges for the reagents used at Rex are as follows:

0-6 years old:	0-60 units
6-10 years old:	0-170 units
adults:	0-85 units

Both of these serological tests provide an aid to diagnosis but must be interpreted in the light of the available clinical evidence and should not be considered diagnostic in themselves.

Karl T. Kleeman, Ph.D.

[1] *Manual of Clinical Laboratory Immunology,* ASM Press, 1992
[2] *Principle and Practice of Infectious Diseases,* Mandell, Bennett & Dolin, 1995.

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