NEWS FROM THE SASKATCHEWAN DISEASE CONTROL LABORATORY

Highlights of This Newsletter:
See page 3 for new syphilis testing algorithm.

See page 5 for update on metallo-
\[-lactamases in Enterobactereae.\]

Update on HIV Point- of-Care (POC) Testing:
The HIV Point-of-Care (POC) testing initiative across the province commenced last spring. The initial phase of the program was implemented in selected hospitals and clinics in Saskatchewan using the INSTI kit from BioLytical with venous blood samples. For 2011 the Ministry of Health will again purchase one lot number for the province. Using a single lot number is an important aspect of quality assurance for jurisdictions that are low volume testing sites. The significant changes for 2011 are: the new kits can be used for finger stick samples, and labs no longer need to provide site ID numbers on samples submitted to SDCL. The HIV POC testing targets four populations:

- pregnant women near term or in labour who have not had a previous HIV test, or have had a recent test and are at risk for HIV.
- in cases of blood/body fluid exposures, testing of the source patient to determine if the exposed person should be offered prophylaxis/treatment.
- acutely ill patients who would be treated with different medications if HIV positive.
- individuals who are at high risk for HIV or whose HIV status is unknown.

For the HIV POC testing, all positive samples will be sent to Saskatchewan Disease Control Laboratory (SDCL) for confirmation (3mL of serum will be required). All low volume HIV POC sites will send both positive and negative samples to SDCL for routine HIV testing. The kits and control materials will be distributed through the Materials Management at SDCL. This will ensure that all sites use the same lot number. Wet lab training for the new kit will be offered in December, 2010 and January 2011. The present HIV POC sites will be notified shortly with the time and location of the wet labs.

Two updated information packages:
(i) a tear sheet titled, HIV Point of Care Kits for Health Care Workers, and

For additional information on the program call the Saskatchewan Ministry of Health HIV/BBP/IDU Consultant at (306) 787-7260.

SDCL New Address:
5 Research Drive, Regina, SK
S4S 0A4

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Core Lab - New Requisition:

In the past few months, many changes have occurred at the Saskatchewan Disease Control Laboratory (SDCL). As we become settled into our new facility at 5 Research Drive, we are evaluating the services we provide our clients and how we can improve the efficiency of these services.

We have recently implemented a Core Lab area to deal with high volume chemistry and immunoserology testing. To reflect this change, a new lab testing requisition form for these areas is being implemented in order to make test ordering simpler for our clients. This new requisition is an integration of our previous testing requisitions.

We will be distributing these new requisitions to our client sites shortly. Upon receipt, please collect and return all copies of the following requisition forms to SDCL:

- Thyroid Function - beige requisition (Health 13-41 06/09)
- Chemistry - green requisition (Health 13-126 09/09)
- Viral Serology - light blue requisition (Health 13-18 02/08)
- Immunoserology - pink requisition (Health 13-104 08/09)
- Syphilis/Rubella/Prenatal - purple requisition (Health 13-2 01/08 rev)
- HIV 1 & 2 Serology (can still be used in the case of HIV POC testing) - blue requisition (Health 13 10/09)

Please review both the front and back of this new requisition carefully, and note the following changes:

- Two areas on the requisition (upper right side) are marked “SDCL Use Only”. Please do not cover these sections with information, as they will be used for accessioning samples at SDCL.
- The patient’s personal health number (PHN) and birth date are used to identify specimens. Please ensure that this information is always included.
- Results will be sent directly to the physician’s address indicated in the “Return Address” box on the front of the requisition.
- Collection requirements for some tests are listed on the reverse side of the requisition. If the submission requirements are not listed, please refer to our SDCL compendium at: http://www.health.gov.sk.ca/compendium

- For RBC folate testing requests please include a copy of the patients CBC result.
- “HIV Confirmation” is a test that can be requested in cases where clients have already performed an HIV screening test on-site (this applies to 3rd and 4th generation EIAs and not to HIV point-of-care testing) and require confirmation of a preliminary screen result.
- Viral load testing is only validated for monitoring therapeutic response to anti-viral drugs in an individual that has already tested positive for hepatitis B, hepatitis C, or HIV and is not used for diagnostic screening purposes.
- For viral serology testing, please indicate if your patient requires an immune status check (check off the IgG box) or if the patient is symptomatic and/or requires testing for an acute infection (check off the IgM box).

For clients participating in the HIV Point-of-Care testing program, all required information can be captured on the reverse side of the requisition. Please include “applicable risk factors” for testing. The separate HIV 1 & 2 Serology requisition (Health 13 10/09) can also still be used.

If you have not received the new Chemistry/Immunology requisition, please order them using the Supply Requisition and fax it to our Materials Management Department at fax number (306)798-0071.
HIV, Hepatitis C and Hepatitis B Viral Load Testing:

In May of 2010 SDCL moved HIV, HCV and HBV viral load testing to a new platform. The new method, Roche COBAS Ampliprep/COBAS TaqMan replaced the Abbott realtime nucleic acid test (NAT) assay.

In the new HIV viral load assay, the assay range for HIV remains the same, 40 - 10,000,000 copies/mL. HIV results will be reported in three formats: (i) HIV RNA not detected; (ii) HIV < 40 copies/mL – RNA detected below reproducible limit of quantification for this assay; (iii) HIV RNA detected, (numerical value) copies per mL.

The new assay range for HCV viral load is 43 to 69,000,000 IU/mL. HCV results will be reported in three formats: (1) HCV RNA not detected; (2) HCV <43 IU/mL – RNA detected below reproducible limit of quantification for this assay; or (3) HCV RNA detected, (numerical value) IU/mL.

The new assay range for HBV viral load is 54 to 110,000,000 IU/mL. HBV results will be reported in three formats: (1) HBV DNA not detected; (2) HBV <54 IU/mL - DNA detected below reproducible limit of quantification for this assay; or (3) HBV DNA detected, (numerical value) IU/mL. The HBV viral load conversion factor for IU/mL to copies/mL is 5.82. Therefore the sensitivity is 54 IU/mL x 5.82 = 314 copies/mL.

Collect blood in vacutainer containing EDTA (lavender stopper) as an anticoagulant. Centrifuge. Aliquot required amount of plasma (4 mL) equally between two (2) 12 x 75 mm plastic tubes as soon as possible. Freeze specimens as soon as possible. If specimens thaw, sample integrity may be compromised for this assay.

Syphilis Testing:

The SDCL’s syphilis testing algorithm will change in November 2010. The new platform will continue to use an initial EIA screen, followed by another confirmation test (TPPA) and a non-treponemal test (RPR). Reactive RPR results are further titrated, to give therapeutic information. While most samples will be reported with initial IgG treponemal antibody screen, IgM treponemal testing will be performed under the following circumstances:

- all prenatal screening
- samples flagged primary syphilis or chancre
- an outbreak investigation
- serum samples from all infants <18 months of age

All samples reactive by IgM and all samples reactive by IgG with either TPPA or RPR reactive are reported to the regional Medical Health Officer.

A four-fold rise or fall in RPR titre is consistent with active syphilis. The RPR cannot be used to diagnose neurosyphilis. To rule out neurosyphilis in symptomatic patients, the best test is CSF for VDRL. Syphilis treatment (benzathine penicillin IM) is available from public health.

For further information regarding this new test, or our testing algorithm, please contact Jim Putz, Section Manager of Immunoserology, at (306) 787-4948.
Guidelines for Ordering Vitamin D Test:
The significance of vitamin D in maintaining good health has received a great deal of attention in recent years.

Serum 25-hydroxy-vitamin D is the analyte of choice for assessment of a patient’s vitamin D level. It is preferred because it reflects precursor levels of vitamin D derived from cutaneous metabolism as well as from dietary intake. The SDCL measures 25-OH-Vit D3, 25-OH-Vit D2 and total 25-OH Vit D (D2 +D3).

The average Canadian’s exposure to sunlight is generally inadequate to maintain vitamin D sufficiency.

Experts advocate that with the recognition of widespread deficiency/insufficiency in children and adults, there is no need to measure everybody’s blood 25 hydroxy vitamin D, rather, asymptomatic, at-risk individuals such as elderly or infirm patients, or those who are believed to receive inadequate sun exposure, receive vitamin supplements as per Health Canada guidelines1. See Health Canada recommendations (http://www.hc-sc.gc.ca/fn-an/food-guide-aliment/context/evid-fond/vita_d-eng.php).

Measurement of serum 25-hydroxy vitamin D may be of clinical value in individuals with conditions in which vitamin D is implicated such as:

- Significant renal or liver disease
- Osteomalacia, osteopenia or osteoporosis
- Possible cases of rickets
- Malabsorption syndromes
- Hypo or hypercalcemia/ hyperphosphatemia
- Hypo or hyperparathyroidism patients on medications that affect vitamin D metabolism such as phenobarbital, carbamazepine, phenytoin and valproate.

- Patients taking high does of vitamin D (>2,000 IU daily) for > 6 months, and who are exhibiting symptoms suggestive of vitamin D toxicity - pain, muscle weakness, vomiting, and/or confusion.

Vitamin D supplementation may provoke hypercalcemia in patients with certain clinical conditions such as parathyroid disease, granulomatous disease, sarcoidosis, lymphoma and kidney disease. The resulting hypercalcemia may increase the toxicity of medications such as digoxin and thiazide diuretics.

Measurement of serum 1,25 dihydroxy vitamin D levels should be limited to patients with significant chronic renal failure (i.e., stage 4 or 5 chronic kidney disease) suspected of having renal 1 alpha hydroxylase deficiency. This test is restricted to specialists.

If measurements of 25-hydroxy vitamin D are undertaken in moderate risk patients, then this testing should follow 3-6 months of adequate supplementation to establish proper levels have been obtained2.

In summary, measurement of serum 25-hydroxy vitamin D levels within a healthy population has little diagnostic value, since supplementation is generally indicated regardless of the outcome of the 25-hydroxy vitamin D test.

References:
Metallo-β-lactamases in Enterobacteriaceae:

Metallo-β-lactamases are enzymes that confer resistance to carbapenem antibiotics (imipenem, meropenem, ertapenem). Initially, these enzymes were restricted to strains of Pseudomonas and Acinetobacter, but metallo-β-lactamases have also occurred in Klebsiella pneumoniae. These enzymes, known as K. pneumoniae carbapenemases (KPC), have also been detected in Escherichia coli.

Carbapenemases are significant because they confer resistance to all β-lactam antibiotics. In addition, carbapenemases found in the Enterobacteriaceae are often carried on plasmids that carry genes for resistance to multiple antibiotics. Such strains are often susceptible only to colistin and tigecycline (Toye et al., 2009).

Recently, there has been much publicity following the detection of other metallo-β-lactamases, known as VIM and NDM, in isolates of K. pneumoniae, E. coli, Enterobacter and Morganella morganii. In all cases reported to date, there has been a history both of travel to Greece (VIM) or India or Pakistan (NDM) and exposure to hospital environments (Deshpande et al., 2010; Kumarasamy et al., 2010; Vatopoulos, 2008). It is important to note that the different metallo-β-lactamases produce phenotypically identical resistance; infection control guidelines for carbapenem-resistant Gram-negative rods have been published recently (CDC, 2009).

Detection of carbapenemase production is complicated because some carbapenemase-producing isolates demonstrate elevated, but susceptible, carbapenem MICs. CLSI has published guidelines for detection of isolates producing carbapenemases (CLSI, 2010). For isolates that test susceptible to a carbapenem but demonstrate reduced susceptibility either by disk diffusion or MIC testing, performing a phenotypic test for carbapenemase activity, the Modified Hodge Test (MHT), is recommended (Anderson et al., 2007).

In Saskatchewan, all isolates of Enterobacteriaceae that are resistant to carbapenems should be referred to the SDCL for confirmation. Isolates with confirmed resistance are forwarded to the National Microbiology Laboratory for molecular identification of the specific metallo-β-lactamases.

References:

In November 2010, the Antinuclear Antibody cascade will change:

Autoimmune disease is characterized by the development of autoantibodies against specific self-antigens. While these autoantibodies may or may not be involved in pathology, they are often useful diagnostic markers for a given disorder. Antinuclear antibodies (ANA) are characteristic of the systemic rheumatic diseases and testing for this family of antibodies is indicated for the evaluation of patients with suspected systemic rheumatic disease and for monitoring patients with evolving or established conditions. A negative result means the patient’s serum shows no reactivity for the most common clinically relevant antibodies associated with connective tissue diseases.

The new platform used at SDCL is Bioplex ANA screen. If the ANA screen is positive, the following analytes are tested:

- Anti-ds DNA (quantitative)
- Anti-Chromatin
- Anti-Ribosomal Protein
- Anti-SS-A (52 and 60)
- Anti-SS-B
- Anti-Smith (Sm)
- Anti-Sm/RNP
- Anti-RNP (A and 68)
- Anti-Scl-70
- Anti-Jo-1
- Anti-Centromere B

Benefits of the new format:

- Detects the most common clinically relevant auto antibody associated with connective tissue diseases.
- Provides greater specificity compared to IFA.
- Determines ANA antigens specificity of specimen found positive using the initial screening sample.
- When anti–double stranded DNA is positive, it will include a semi-quantitative result.
- Comes with medical decision support software (MDSS) algorithm that provides a suggested disease association based on a database on how individuals with similar autoantibody values had been diagnosed.

For example, in one study the medical decision support software (MDSS) algorithm suggested an appropriate disease association in 75 to 100% of patients with SLE. In summary, each pattern is analyzed by an interpretative algorithm, and a suggested disease association provided.

The Hep 2 IFA (ANA Hep 2 IFA) is available upon a phone request by physician to (306) 787-4948. This test is more sensitive, but has a lower specificity for lupus.

The antinuclear antibody cascade cannot be used to identify rheumatoid arthritis. Up to 50% of patients with rheumatoid arthritis can be positive for ANA. To rule out RA add the rheumatoid arthritis markers RF (rheumatoid factor) and CCP (cyclic citrullinated peptide).

For further information regarding this test, please contact Jim Putz, Section Manager of Immunoserology, at (306) 787-4948.
Healthcare Provider Information:
To reduce processing delays and the number of phone calls to your facility, please ensure complete and accurate healthcare provider information is included on all requisitions or packing slips that you send to SDCL:

Physician or Nurse Practitioner’s Last Name, First Name or initials as it appears in the Saskatchewan Physician Registry MUST be included on all requisitions.

- For identity purposes, also include Physician’s or Nurse Practitioner MCIB or Billing Number
- When using an out of province physician as an ordering doctor, provide name of city, province

Do not use:
- Interns or residents as the ordering physician
- Abbreviated last names or ‘Nick’ names for healthcare providers
- RNs unless they have a nurse practitioner designation

CMV PCR or Quantitative CMV PCR Reminder:
On June 1, 2010, SDCL started sending the Quantitative CMV (Cytomegalovirus) PCR testing to Saskatoon Health Region. Please continue to collect and ship specimens as per current practice by sending EDTA anticoagulated blood (one 5ml lavender top tube). Ensure the blood is kept cold (not frozen) after collection and immediately ship to SDCL. SDCL will separate the plasma and refer the specimen on to Saskatoon. If the specimen cannot be sent the same day to SDCL, the specimen must be poured off the clot. As per instructions in SDCL’s compendium, prior to shipping the CMV PCR or Quantitative CMV PCR sample to SDCL, please call 787-3131 Ext. 3 for Out of Country or Province Referrals and speak directly to our Referral staff.

Group Specimens Together for the Same Patient:
In an effort to improve efficiency, and reduce processing delays, SDCL would like to remind clients to please group specimens together for the same patient in the totes. For example, multiple serum specimens (chemistry and immunology test orders) for patient “Jane Doe”, can be grouped together in a row for that patient in one area of the foam rack. Paper clip the corresponding requisitions for the same patient in order of the specimens in the foam racks.

SDCL Tote Biological Specimen Transport guidelines are available upon request.

If you have any questions specific to our Specimen Management Centre processes, please call Darlene Miller, Section Manager at 306-787-3238.

REMEMBER:
SDCL’s New Address:
5 Research Drive
Regina, SK
S4S 0A4
The Saskatchewan Disease Control Laboratory (SDCL) phone tree (306) 787-3131 has been created to help assist with timely response to client inquiries. Provided is a list of the tree numbers and information that clients will access when choosing a number.

**Hours of Operation:** 8:00 a.m. to 5:00 p.m. Monday to Friday
7:30 a.m. to 4:00 p.m. Saturday
Evenings and Weekend Emergency and After Hours Service, see #8 for cell numbers.

**Press 1:** Test Results or to arrange STAT Testing

- Identify who you are, institution and if you are calling for:
  a) Lab results;
  b) Specimen receipt and/or status;
  c) STAT testing (organ donors or needle stick exposure), call will be transferred to the appropriate section; or
  d) Requests for technical information, call will be transferred to appropriate section.

**Press 2:** Compendium Information (listen only) - To access the on-line compendium go to www.health.gov.sk.ca/compendium.

**Press 3:** Out of Province or Country Referrals - For information on tests referred out-of-province or country.

**Press 4:** Exceptions to Provide Missing Information - To provide missing specimen information that has been requested by SDCL staff.

**Press 5:** Maternal Serum Screening - For results on maternal serum screening samples submitted.

**Press 6:** Water Samples - For results on water samples submitted.

**Press 7:** Shipping and Supplies - For example, requests for requisitions or transport media such as SAF, Carey Blair or viral for specimens submitted to the Saskatchewan Disease Control Laboratory.

**Press 8:** Emergency and After Hours Service (Evenings and Weekend) (listen only)

  - STAT Toxicology 536-4653
  - STAT Serology/Microbiology/Virology 537-0639

If you wish to repeat this menu please stay on the line.

**Paper Requests:**

1. Laboratory does not accept verbal test requests and requires you to fax in a request/requisition for any additional testing.

2. If patient cannot be identified, as in “circle of care” then we require a “signed release of medical information form” faxed to us at (306) 787-9122.

**REMEMBER Call (306) 787-3131 for information from the Saskatchewan Disease Control Laboratory.**
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Laboratory Evidence of Human Viral Infections - # of Positive Findings 2010
Please review the protocol for RBC Folate testing

Please note:
The sample must arrive at SDCL ASAP as analysis is only valid for 4 - 5 days after collection

**Section:** Chemistry

**Test:** RBC Folate

**Specimen:** 1 mL Whole Blood

**Collection:** Whole Blood (EDTA): Collect blood after overnight fast (minimum 12 hours). Collect whole blood in vacutainer containing EDTA (lavender stopper). Gently invert specimen a couple of times to prevent clotting. Record fasting, date and time of collection.

**Special Handling Instructions:** NOTE: RBC Folate testing will be done ONLY if the CBC is abnormal or if patient has neurological or other symptoms suggestive of folate deficiency. This information MUST be supplied on the requisition or sample will not be tested.

NOTE: CBC including Hematocrit MUST be supplied. Staple CBC report to RBC Folate order.

**Shipping:** Package as required by transport system used. Ship in accordance with Transport Canada Regulations.

**Reference Values:** 630 - 1,800 nmol/L

**Turnaround Time:** 3 days
**Supply Requisition**

5 Research Drive, Regina, SK S4S 0A4

**Shipping Department Direct Line:**
Phone: 787-3192
[For Supplies only] Fax: 798-0071

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**Date of Order**

**Ordered by**

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**Ship To**

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**Phone #**

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Please print clearly.

Specimen containers with media should be refrigerated (not frozen). Shelf life is limited, so please limit orders to a maximum of 30 days supply.

Non-perishables should be limited to 90 days supply.

Please return any unwanted supplies.

**NOTE:** Supplies ordered from the Provincial Laboratory are only to be used for specimens returning to the Provincial Laboratory for testing.

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### Requisition Forms For:
- Pads/100
- Chemistry and Immunoserology
- Chlamydia
- HIV 1&2 Serology
- Maternal Serum Screen
- Microbiology (Bacteriology)
- Mycology
- Severe Respiratory Illness
- Virology (Culture)

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**Explanation Codes For Order Adjustments**

R.O. - Reduced Order - Your order appears high compared to the number of specimens normally submitted to the Lab for 90 (or 30) days and has been reduced accordingly. If we have misjudged your need, please reorder with clarification.

---

**Date Shipped**

**Invoice #**

**Shipped By**