Sputum Sample Collection for Tuberculosis

A good specimen = better results

TB is caused by *Mycobacterium tuberculosis* (MTB), an extremely slowly-growing bacterium. Laboratory culture of specimens for MTB is challenging and measures are necessary to avoid a delay in laboratory diagnosis or overgrowth of normal flora bacteria present.

Reliable results depend on specimen quality. An optimal volume of 5 to 10 mLs (or a minimum of 1 tsp.) of sputum (not saliva) from a symptomatic patient provides the best chance at successful detection and isolation of MTB from a clinical specimen, particularly in smear negative cases. Insufficient specimen quantity can result in a false-negative result and/or delay in detection. Specimen should be sent to the laboratory as soon as possible and batching should be avoided. If delivery to the laboratory is delayed > 1 hour, refrigeration at 4°C is required prior to transport.

For further information on sputum specimen collection or other specimen types, please refer to the SDCL compendium of tests, under Mycobacterium Culture (TB).

The SDCL Compendium of Tests, requisition pdfs and shipping instructions are available on-line at [http://sdcl-testviewer.ehealthsask.ca](http://sdcl-testviewer.ehealthsask.ca)
Chlamydia is a genital infection caused by the bacterium *Chlamydia trachomatis* and is the most commonly reported sexually transmitted infection (STI) in Canada. Infections are often asymptomatic in both males and females. In the absence of screening, these infections remain undiagnosed and contribute to the spread of chlamydia in sexually active individuals. A common complication associated with untreated and recurring chlamydia in females is pelvic inflammatory disease, which can lead to chronic pelvic pain, ectopic pregnancy and infertility. In males, complications are rarer but include epididymo-orchitis and infertility. Untreated chlamydia in pregnant women can be transmitted to their newborns, causing neonatal conjunctivitis or pneumonia. As with other STIs, chlamydia increases infection with and transmission of the human immunodeficiency virus (HIV). It recruits target cells for HIV to the genital tract and increases the shedding of HIV-infected cells 1, 2.

Between 1991 and 1997, the rate of reported cases of chlamydia decreased steadily among both males and females, after which rates began to rise 3. There are, however, several factors that must be taken into account when considering these data. The introduction of more sensitive nucleic acid amplification testing (NAAT) in the mid-1990s undoubtedly led to an increase in the number of chlamydia cases detected. NAAT allows urine specimens to be used rather than swabs, which are easier to collect and more acceptable to patients. As a result, the number of people, particularly males, who go for testing, has likely increased as well. More effective screening and contact tracing may have a similar effect 4, 5. A recent estimation of chlamydia disease burden in Canada found that observed increases in chlamydia prevalence could be explained by effective case finding and expansion of screening programs 6.

Chlamydia culture on cervix, urethra and eye specimens has been discontinued and been replaced by the more sensitive NAAT. For your information, Chlamydia culture on cervix, urethra and eye specimens has been discontinued and been replaced by the more sensitive NAAT. Please use the APTIMA Unisex Swab Collection Kit (white) for these specimens. NAAT is also performed on vaginal and extra-genital specimens including rectum and throat. Please use the APTIMA Vaginal Swab Collection Kit (orange) for these specimens.

Over time, the rate of reported cases of chlamydia has consistently been higher in females than in males; however, this disparity is much more pronounced in younger age groups. Younger women are biologically more susceptible to chlamydial infection 7, 8. In addition, women are more likely to be screened for STIs 9, 10.

Gonorrhea has been nationally notifiable since 1924 and is the second most commonly reported STI in Canada. Gonorrhea is a genital bacterial infection caused by *Neisseria gonorrhoeae* and if left untreated, can lead to complications for both sexes. There can be severe consequences for females, including pelvic inflammatory disease, which often leads to chronic abdominal pain, infertility and ectopic pregnancy. In males, untreated infections can result in epididymitis and rare cases of infertility. An uncommon complication of gonorrhea is the spread of infection to the blood stream and joints 11. Like other STIs, gonorrhea increases the risk of HIV acquisition and transmission, possibly by increasing the concentration of HIV target cells in genital secretions and viral shedding 2.

Last year, (2014), SDCL ran 80,978 chlamydia and gonorrhea NAAT tests — 57,970 female and 23,008 male. For chlamydia the positive rate for females was 6.7% while the positive rate for males was 9.0%. For gonorrhea the positive rate for females was 1.2% and the positive rate for males was 2.7%.
References:


2. **Fleming DT, Wasserheit JN.** From epidemiological synergy to public health policy and practice: The contribution of other sexually transmitted diseases to sexual transmission of HIV infection. Sex Transm Infect. 1999 Feb;75(1):3-17.


To receive future copies of the SDCL Newsletter electronically and to save trees, please email: Janette Romanuik at janette.romanuik@health.gov.sk.ca with your email address, organization, address and phone number.
Whooping Cough by PCR

On the SDCL Microbiology Requisition indicates two choices for detecting pertussis in nasopharyngeal swabs: Bordella Screen and Pertussis Culture.

The **Bordetella Screen** is a sensitive nucleic acid amplification-based (PCR) test that detects and differentiates DNA from *Bordetella pertussis*, *B. parapertussis*, and *B. holmesii*.

Nasopharyngeal swabs submitted in UTM will receive the **Bordetella Screen**, but are also tested simultaneously for multiple respiratory pathogens as part of the **Respiratory Screen**.

Nasopharyngeal swabs intended for **Pertussis culture** must be submitted in Regan-Lower medium. This specimen will also receive **Bordetella Screen** in addition to culture.

Culture is less sensitive than PCR; many samples (73%) are found to be culture negative and PCR positive. However, if culture is successful, this method provides isolates for surveillance.

Updates for Screening & Reference

**C-reactive Protein (CRP):**

CRP is an acute phase reactant, a protein made by the liver and released into the bloodstream within a few hours after tissue injury, the start of an infection, or other cause of inflammation. SDCL has recently adjusted the reference range for this test to more accurately define normal versus abnormal ranges.

Reference ranges:
- < 8 mg/L: Normal
- >10 mg/L: Suggests inflammation and/or infection

**High sensitivity C-reactive Protein (hsCRP):**

A more sensitive CRP test, called a high-sensitivity C-reactive protein (hsCRP) assay, is available to determine a person’s risk for heart disease. If clinical assessment of cardiovascular risk is required, hsCRP should be ordered. SDCL has recently adjusted the reference ranges for this test to more accurately define standardized levels of cardiac risk.

Risk for cardiovascular disease:
- Low risk: < 1.0 mg/L
- Medium risk: 1.0 – 3.0 mg/L
- High risk: > 3.0 mg/L

**Red Blood Cell Folate Testing:**

A copy of the CBC report must be attached to the order requisition for RBC folate requests. In the future, the responsibility will be on the submitting client to provide this report or the testing may not be performed.

Currently RBC folate testing is only performed on patients that have abnormal hematology results or neurological symptoms. Either an elevated MCV (mean corpuscular volume) and/or low hemoglobin level is required before SDCL will perform RBC folate testing. In this regard, it is imperative that these abnormal CBC (complete blood count) results accompany all requisition orders for RBC folate analysis. Laboratory staff spends significant amounts of non-value added time calling facilities to obtain this information.
**Hepatitis A testing Algorithm:**

In March 2015, SDCL discontinued testing for Total Hepatitis A Antibody replacing it with specific testing for Hepatitis A IgG and Hepatitis A IgM. If a clinician is testing for immune status they should order Hepatitis A IgG, conversely if a clinician is testing for an acute infection they should order Hepatitis A IgM. For acute hepatitis work-up choose “hepatitis screen” that includes hepatitis panel containing hepatitis A IgM, hepatitis B surface antigen, hepatitis B core total and Hepatitis C antibody. Please use the SDCL requisition form Doc 1400-55 SDCL003R when ordering hepatitis tests.

**Vitamin D:**

Effective April 1, 2015, SDCL will offer only one option for analysis of Vitamin D levels; 25-OH Vitamin D Status (Combined Total D2 & D3).

**Total protein analysis on urine is no longer available at SDCL.**

This test is still available at Regina General Hospital (RGH). Please refer to the RGH compendium of tests for sample collection/submission requirements. Please send these requests directly to RGH.

**Hepatitis C Diagnostic Testing at SDCL:**

Hepatitis C virus (HCV) is an RNA virus that belongs to the family *Flaviviridae*. The virus was first identified in 1989 using molecular methods \(^1\). HCV is a global health care problem; chronic infection has been estimated to affect up to 1% of the population in Canada \(^2\) and may lead to fibrosis, cirrhosis and hepatocellular carcinoma \(^1\). Most acute infections with hepatitis C are asymptomatic. \(^3\)

Laboratory diagnosis of HCV is a multi-step process, in which antibodies are detected using a screening test, followed by a more specific supplemental or confirmatory test. This approach prevents reporting of false-positive results, particularly in settings where asymptomatic persons are being tested \(^4\)\(^5\). However, the presence of HCV antibodies cannot distinguish between individuals whose past HCV infection has resolved and those who have active infection \(^6\). Identification of active infection allows for targeted follow-up of patients who may benefit from treatment. Active infection can be confirmed by the detection of HCV RNA (by PCR) or by HCV core antigen detection \(^7\)\(^9\).

Detection of viral RNA by PCR requires the collection of an additional plasma sample, which delays the completion of testing and may also result in patients being incompletely diagnosed \(^10\), and is also relatively expensive compared to other diagnostic tests. The recent availability of a sensitive assay for HCV core antigen allows for completion of diagnostic testing using the initial serum sample \(^9\).

HCV antigen tests have recently been incorporated into the algorithm for hepatitis C diagnostic testing at SDCL. Serum specimens that are reactive in the HCV antibody test will be tested for HCV antigen.
A reactive antigen test leads to a report that the specimen tested positive and represents active infection.

A non-reactive antigen test will receive further testing to confirm the presence of HCV antibodies and a report that the patient may require further testing, depending on symptoms and risk-factors.

Further testing available at SDCL includes HCV viral load assays, HCV genotyping and Q80K polymorphism testing of genotype 1a strains (the latter is a referral test to the BC Centre for Excellence in HIV/AIDS). The application of these tests to support the treatment of HCV infection will be described in the next issue of the newsletter.

References:


Reminder: A complete mailing address or validated fax number is required for all copy-to requests. Failure to submit this information will delay in you receiving results for your patient.
Monitoring Exposure to Blood and Body Fluids:

When the source’s (patient) results are unknown regional laboratories can request a STAT work-up for “needle stick exposure”. This STAT testing is to be pre-arranged with SDCL. The client requesting this service must phone and arrange approval from the section or after hours with the microbiologist-on-call. STAT testing is performed as soon as possible after arrival of the sample at SDCL.

During regular hours of operation, 8:00 a.m. to 5:00 p.m. Monday to Friday and 7:30 a.m. to 4:00 p.m. Saturday, phone (306) 787-3131, option #1, identify as “needle stick exposure” STAT and the call will be transferred. After hours, evenings and weekends, phone (306) 798-1234, the microbiologist-on-call to make arrangements.

In your communication (phone call) of STAT request, include the following information:

- Mode of transportation – courier, bus, cab, etc.
- Date and time sample will be sent out and expected time of arrival in Regina.
- Patient information: name, date of birth, health services number (HSN).
- Tests requested – HbsAg, anti-HBs, HB core total, HCV and HIV.
- Name of facility sending sample.
- Name of contact, contact’s phone and fax numbers as it may be necessary to call the contact if the sample does not arrive at expected time.
- Name and contact phone number of the person to receive the result.

When using the SDCL Chemistry & Immunoserology Requisition Doc 1400-55 SDCL003R, check the “needle stick” work-up box. This will trigger the testing panel that includes HBsAg, anti-HBs, HB core total, HCV antibody and HIV antibody tests. Include the patient’s name and another unique identifier such as HSN. Mark “STAT” on the requisition; label the sample (tube) with two unique identifiers and the initials of the person who took the sample. When using a manifest print, please indicate “needle stick exposure”.

Samples are to be placed in plastic Ziploc bag clearly marked “STAT” which can be shipped with routine samples, and inside a tote flagged with a “yellow” closure tie. If using Saskatchewan Transport Company (STC) for a “STAT” delivery, mark the manifest “Station to Station” for quickest delivery time.

A Change in Reporting Rubella Immunity:

The SDCL is now reporting rubella titres between 10 and 15 IU/mL as indeterminate and these individuals should be considered non-immune.

The change in reporting is based on a review of laboratory data which indicates that levels of rubella antibody close to the break point of 10 IU/mL fluctuate above and below this value on repeat testing. Therefore, samples between 10 and 15 IU/mL are better classified as indeterminate.
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: Monday to Friday 8:00 AM to 5:00 PM.
On Saturdays limited services 7:30 AM to 4:00 PM.

**Press 1: For general medical laboratory information, test results and to arrange for Stat testing**

Customer Services staff provides information to callers and responds to requests for lab test results. If all lines are busy, you may leave a message (provide details) for a return call or to have lab test results faxed (validated fax number only).

For lab test results, provide the patient HSN, test, collection date, validated fax number, who is calling and return phone number.

**Note** for STAT testing or requests for technical information, your call will be transferred to the appropriate lab section.

**Press 2: Compendium Information** - To access the on-line compendium go to http://sdcl-testviewer.ehealthsask.ca

**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 from requisitions sent to SDCL** - To provide missing sample 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, Cary Blair or viral for samples submitted to the Saskatchewan Disease Control Laboratory.

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

  - STAT Toxicology (306) 536-4653
  - STAT Serology, Microbiology or Virology (306) 537-0639

Note for Call backs: Provide the patient’s HSN, test required, name of physician requesting the STAT testing.

Or stay on the line to repeat the menu.

**Requests for Add-on Tests or Laboratory Results (Outside “Circle of Care”)**:

1. SDCL does not accept verbal requests for testing and requires a faxed (or hard copy) request (or requisition be submitted for add-on test requests.

If a request for laboratory results for a patient is made by someone not obviously identified as in the "circle of care", before release of results can occur, SDCL requires a "signed release of medical information form" to be faxed to (306) 787-9122. For a copy of the release form, contact SDCL at (306) 787-3131.