



LABORATORY DIAGNOSIS OF SEXUALLY TRANSMITTED INFECTIONS

This chapter from the Canadian Guidelines on Sexually Transmitted Infections 2006 Edition has undergone revisions and has been updated as of October 2007. The chart below summarizes the most significant changes made to the chapter and cross-references the corresponding page numbers in the current hard copy version of the guidelines.

<u>Section</u>	<u>Page</u>	<u>Current Wording/Problem</u>	<u>Update/Clarification</u>
C. Laboratory diagnosis of specific infections 1. Chlamydia trachomatis	35	Need to strengthen/clarify statements around the use of serologic testing for the diagnosis of Lymphogranuloma Venereum (LGV).	Addition to last bullet Due to issues of cross-reactivity and difficulty with interpretation of test results, serological testing should not be used for diagnostic purposes in the absence of culture or NAAT.
2. Neisseria gonorrhoea	36	NAAT can be used to detect reinfection after waiting for at least 2 weeks after completion of therapy.	NAAT can be used to detect reinfection after waiting for at least 3 weeks after completion of therapy.
5. Treponema pallidum	37	Guidance on the use of EIA for diagnosis of syphilis	Bullet added: The introduction of treponemal tests for IgG/IgM antibodies, such as the treponemal enzyme immunoassay (EIA), may provide a more sensitive screening test for syphilis. Although EIA is highly sensitive, the test can lack specificity therefore if the treponemal-specific ELISA is positive, confirmation by a second treponemal-specific test is required (e.g. TP-PA, MHA-TP, FTA-ABS).

LABORATORY DIAGNOSIS OF SEXUALLY TRANSMITTED INFECTIONS

A. COLLECTION AND TRANSPORTATION OF SPECIMENS¹

General principles

- Swabs, transport systems and types of tests used may vary depending on the agent sought and techniques used by the laboratory.
- Contact the laboratory to obtain further information, especially concerning transport requirements, turn-around time and interpretation of results. See *Appendix E* for a listing of local contact information.
- Laboratories may use a variety of commercial specimen-collection devices. Follow the instructions provided by the manufacturer.
- All specimen-collection and handling procedures should be performed while wearing appropriate protective clothing and following recommended universal precautions.
- Contamination from indigenous commensal flora should be avoided to ensure a representative sampling of organisms involved in the infectious process.
- Adequate volumes of each liquid specimen should be collected.
- Each specimen container should be labelled with the patient's name and identification number, the source of the specimen and the date and time of collection.
- All specimen containers should be leak-proof and transported within a sealable, leak-proof plastic bag that has a separate compartment for paperwork.
- Sexually transmitted pathogens are usually fastidious and fragile; cultures and techniques that detect viable organisms may give false-negative results unless storage and transport conditions are optimal.
- Storage recommendations need to be observed, and transport must be as rapid as possible for the recovery of infectious organisms, with excesses of temperature avoided.

Specimens

For most sexually transmitted infections (STIs), specimens will be collected by health care providers to be packaged and delivered to diagnostic laboratories. There is an effort to produce commercial point-of-care testing kits for in-office testing, but there are none that are approved and validated at this time. Self-collection of urine, vaginal and lesion swabs is currently being evaluated for home collection, but these strategies lack appropriate evaluation, especially for transportation conditions.

1. *Cervix*

- After inserting a speculum to view the cervix, remove overlying vaginal secretions and cervical exudate.
- Insert a sterile swab 1–2 cm into the endocervical canal, rotate 180° and withdraw for collection of columnar epithelial cells for diagnosis of *Chlamydia trachomatis* and *Neisseria gonorrhoeae*. The choice of swab should be based on the type of testing being done; consult with the laboratory providing the service.
- Obtain a specimen for *N. gonorrhoeae* before taking a specimen for *C. trachomatis*.
- If a culture is to be performed for *N. gonorrhoeae*, directly inoculate the culture tube or plate, or place the swab in the transport medium. Alternatively, place the swab in a nucleic acid amplification transport tube.

Exocervical samples are better for herpes simplex virus (HSV) and human papillomavirus (HPV).:

Notes:

- Cervical specimens should not be taken from prepubertal girls, since STIs in this age group involve the vagina, not the cervix. See *Sexual Abuse in Peripubertal and Prepubertal Children* chapter for more information.
- Obtaining several specimens from the cervix does not usually produce discomfort and may be required to perform various tests.
- In women who have had a hysterectomy collect first void urine or vaginal swab for NAAT*

*Although some NAATs have not been approved in Canada for use with vaginal specimens, recent data show that women infected with *C. trachomatis*, *N. gonorrhoeae* or *Trichomonas vaginalis* may be identified more often using vaginal swabs than cervical swabs, urethral swabs or urine.^{2,3} Check with your laboratory to see if this is an option.

2. Lesions (vesicles or ulcers)

a) Vesicles

- Fluid can be obtained by lifting the top of the vesicle and swabbing the lesion.
- An alternative method is to clean the vesicle with a disinfectant and, after drying, piercing into the fluid with a syringe, collecting fluid, capping, sealing the plunger and transporting to the laboratory.

b) Ulcers

- Warn the patient that specimen collection may be painful.
- Swab the lesion bed for culture, polymerase chain reaction (PCR) or direct examination for HSV.
- For direct examination, obtain cellular material by firm swabbing or gentle scraping from the base of the lesion.
- For culture, use the swab and viral transport medium supplied with the collection kit.
- For the detection of *Treponema pallidum*, contact the laboratory to determine the availability of dark-field microscopy or direct fluorescent antibody (DFA) testing. Where available, collect a specimen as follows:
 - Remove scabs or overlying debris.
 - Cleanse the lesion with sterile saline without preservatives and dry the area.
 - Abrade the lesion with a dry sterile gauze pad to provoke slight bleeding and exudation of tissue fluid.
 - As oozing occurs, wipe away the first few drops and await the appearance of relatively clear serous exudate. It is sometimes necessary to apply pressure at the base of the lesion to express tissue fluid.
 - Collect fluid into a capillary tube, small-bore syringe or directly onto a slide for DFA testing.
 - Seal the tube, cap the syringe or immobilize the plunger before transportation.
 - Store at 4°C before transportation and deliver to the laboratory within 24 hours.

- For *Haemophilus ducreyi*, a special medium is required for culture. Obtain a swab from the base of a lesion, avoiding pus, and place into a transport tube.

3. Pharynx

- Swab the posterior pharynx and the tonsillar crypts.
- Use the swab to directly inoculate the appropriate culture medium, or place it in a transport medium.
- For infants, obtain a nasopharyngeal aspirate.

Notes:

- There are promising data on the performance of non-culture NAAT tests using pharyngeal specimens.
- Smears of pharyngeal swabs are of no value in detecting pharyngeal *N. gonorrhoeae* by microscopy and are not recommended.

4. Rectum

- For blind swabbing, insert 2–3 cm into the anal canal, press laterally to avoid fecal material and, in the case of *C. trachomatis* or *N. gonorrhoeae*, to obtain columnar epithelial cells.
- If there is visible fecal contamination, discard the swab and obtain another specimen.
- With unlubricated anoscopy using only tap water, fecal contamination can be avoided and specimens can be collected under direct visualization.

Notes:

- Specimens may be obtained blindly or through an anoscope. The latter is preferred for symptomatic patients.
- There are promising data on the use of rectal and oral swabs for *C. trachomatis* and *N. gonorrhoeae* tested by NAATs and current clinical trials are underway through the U.S. National Institutes of Health.

5. Urethra

- Warn the patient that specimen collection may be painful, that the next urination may be painful, and that increasing fluid intake may help to decrease urine concentration and therefore discomfort.
- Ideally, the patient should not have voided for at least 2 hours, as voiding reduces the amount of exudate and may decrease the ability to detect organisms.
- Use a thin, dry swab with a flexible wire shaft. Moistening the swab with water before insertion may help reduce discomfort.
- Introduce the swab slowly (3–4 cm in males; 1–2 cm in females), rotate slowly and withdraw gently.
- The swab can be used to prepare a smear by slowly unrolling the secretions onto a slide; then, directly inoculate the appropriate culture medium or place the swab in a transport medium.
- If a NAAT is used, follow the manufacturer's instructions.

Notes:

- “Milking” the penis three or four times from the base to the glans enhances the ability to detect otherwise inapparent urethral discharge.
- In prepubertal boys and girls, collection of an intraurethral specimen is not recommended; obtain first-void urine specimens for NAATs or a meatal specimen using a thin swab with a flexible wire shaft.

6. Urine (first-void)

- The patient should not have voided for at least 2 hours, but having done so does not preclude testing.
- Provide the patient with a leak-proof container.
- Ask the patient to collect *only* the first 10–20 mL of urine^{3,4} into the container and to cap it tightly.

Note: Most commercial NAATs for *C. trachomatis* and *N. gonorrhoeae* are approved for urine testing and are recommended for detecting these organisms in asymptomatic men or women, women without a cervix or those who wish to avoid pelvic examination. A first-void urine (FVU) may be collected at any time and may also be termed a first-catch urine (FCU).

7. Vagina

- Collect pooled vaginal secretions, if present.
- When no vaginal secretions are present, swab the vaginal wall in the posterior fornix to prepare a smear, or place the swab in a transport medium.
- Wet-mount and Gram-stain smears are useful in the diagnosis of microbial vulvovaginitis, candidiasis, bacterial vaginosis, trichomoniasis or desquamative inflammatory vaginitis.
- Collection of vaginal specimens from youth and adults is usually done as part of a speculum examination.

- In prepubertal girls, vaginal-wash specimens are most preferred and patient acceptable. If not possible, use swabs moistened with water. See *Sexual Abuse in Peripubertal and Prepubertal Children* chapter for more information.
- In very young children, use very thin swabs.

Note: In the past, vaginal specimens were not recommended for the diagnosis of STIs, except in the management of vulvovaginitis, bacterial vaginosis and child sexual abuse. More recent data show that NAATs for *C. trachomatis*, *N. gonorrhoeae* and *Trichomonas vaginalis* may identify as many or more infected women using vaginal swabs than cervical swabs, urethral swabs or urine.^{2,3} Check with your laboratory to see if this is an option.

8. Warts and Other HPV Infections

- Scrape the exocervix for superficial epithelial cells.
- Cytobrushes, other collecting devices or swabs can be used to collect cells from the squamo-columnar junction of the cervix.
- Currently commercial and non-commercial assays with specific collection devices are available for HPV DNA detection. Consult with your laboratory.

Note:

Urine samples have not been shown to be as accurate as cervical samples for detecting high-risk HPV.⁵

B. LABORATORY TESTING METHODS

STIs may be diagnosed in the laboratory using (a) culture, (b) microscopy, (c) antigen detection, (d) nucleic acid detection, (e) serology and (f) surrogate markers. The sensitivity and specificity of these different approaches vary according to specimen type and organism assayed. The number of false positives or negatives will be influenced by the prevalence of infection in the population being sampled. NAATs are the most sensitive methods, and culture the most specific. Antigen detection, nucleic acid hybridization, culture and microscopy are less sensitive but may be effective for certain types of patients and specimen types. Since not all diagnostic laboratories perform the same tests, clinical conditions and specimen types should be discussed before collecting the specimen. In some situations, serology is very useful (e.g., syphilis), but in others (e.g., non-LGV *C. trachomatis*) it is of no use. Surrogate markers, such as leukocyte esterase strip tests, pH or amines point-of-care tests may provide useful screening for some conditions, but are generally insensitive and not very specific.^{6,7}

C. LABORATORY DIAGNOSIS OF SPECIFIC INFECTIONS

1. *Chlamydia trachomatis*

- Results are highly dependent on the type of test available,⁸ appropriate specimen collection,⁹ storage, transport and laboratory expertise.
- Contact the laboratory for specific instructions before submitting specimens, and read and follow test-kit instructions regarding specimen collection, storage and transport.
- NAATs are the most sensitive and specific and should be used whenever possible for urine, urethral, and cervical specimens; blood and mucous can affect NAAT performance.³
- Non-invasive specimens such as urine can be used in NAATs, making testing more acceptable to patients.¹⁰
- Both *C. trachomatis* and *N. gonorrhoeae* can be detected from a single specimen in some NAATs.¹¹
- Because successful treatment rates are high, a test of cure is not usually performed.
- Other assays, such as nucleic acid hybridization and antigen detection, may be used, but they are less sensitive and specific, and positives may need to be confirmed.¹²
- *C. trachomatis* IgM serology is useful for diagnosing *C. trachomatis* pneumonia in infants under 3 months of age.¹³
- Serology is not useful for the diagnosis of acute genital chlamydial infections (non-LGV only).
- Culture is the preferred method for medico-legal purposes, but NAATs may be suitable, provided that positive results are confirmed. Confirmation of positive results can be done with a NAAT using a different set of primers or by DNA sequencing techniques.
- Strains of lymphogranuloma venereum (LGV) have emerged in Europe and North America, mainly in rectal samples (RS) of men who have sex with men (MSM). Existing NAATs are not cleared by the U.S. Food and Drug Administration or Health Canada for use on rectal or oropharyngeal samples, but will record positives that need to be confirmed as LGV by restriction fragment length polymorphism (RFLP) or sequencing techniques. Samples can also be cultured undiluted and at a dilution of 1:10 (to dilute fecal toxicity) using shell vials with and without centrifugation. LGV grows readily to high levels of elementary bodies without centrifugation, while non-LGV strains require centrifugation. As with NAATs, positive cultures need to be confirmed as LGV by RFLP or sequencing. A NAAT or culture can also be used on other samples in the diagnosis of LGV such as bubo aspirates; urine; or rectal, vaginal or urethral swabs. Emphasis should be placed on clinical samples for a definitive diagnosis; however, serology such as microimmunofluorescence (MIF) may be helpful in supporting the diagnosis. Due to issues of cross-reactivity and difficulty with interpretation of test results, serology should not be used for diagnostic purposes in the absence of culture or NAAT. For more information on specimen collection and available tests, please contact your local laboratory (see *Lymphogranuloma Venereum* chapter for more information on specimen collection and testing by stage of infection).

2. *Neisseria gonorrhoeae*

- The presence of Gram-negative diplococci inside polymorphonuclear leukocytes (PMNs) is highly predictive for the direct microscopic examination of smears; their presence outside PMNs is not, and confirmation by culture is required.
- The sensitivity and specificity of the Gram stain depends on the type of specimen.¹⁴ Urethral specimens from young adult males have a sensitivity and specificity of 95%; endocervical specimens from adult females have a sensitivity of 45–65% and a specificity of 90%.
- Culture for *N. gonorrhoeae* is required for the determination of antimicrobial susceptibility in cases of sexual abuse/assault, as well as in cases of treatment failure.
- Successful culture of specimens requires proper collection and transportation of appropriate specimens or immediate inoculation of medium.¹⁵ Consult with your laboratory.
- NAATs are approved for cervical and urethral swabs and urine; some NAATs are also approved for vaginal swabs.¹¹ Urine and vaginal swabs are convenient specimens for women without a cervix, and urine may be most convenient for those who may not readily submit to a pelvic examination.
- Urine is a preferred specimen for men if a NAAT is performed.
- A NAAT is not recommended as a test of cure.
- NAAT can be used to detect reinfection, after waiting for at least 3 weeks after completion of therapy.
- For medico-legal purposes, a positive result obtained from NAATs should be confirmed using a different set of primers or by DNA sequencing techniques.
- Serology is not available.

3. *Hemophilus ducreyi* (chancroid)

- Because *H ducreyi* is rare in Canada, consult with your laboratory.
- Culture is the current method of choice, using two media in a biplate.¹⁶
- Specimens of choice are a calcium alginate or cotton swab from the base of the ulcer or an aspirate if buboes are present.
- There are no useful serologic tests to diagnose *H ducreyi*. Gram stain with Gram-negative coccobacilli in a “school of fish” pattern may be useful.
- If a NAAT is available, a second ulcer swab should be collected into an appropriate transport medium.

4. Herpes simplex virus

- NAATs are being used increasingly for cerebrospinal fluid, vesicle fluid or ulcer swabs.¹⁷ Consult with your laboratory.
- NAATs approach sensitivities and specificities of 100%, with rapid turn-around of results.
- Cultures are easy to perform and can yield positive results within 24 hours from primary or first-episode genital herpes.
- Other methods, such as antigen detection and Tzanck smear cytology, lack accuracy.
- For neonates, gently rub conjunctiva, insert separate swab into mouth (and gently rub around the lips), external ear canal, umbilicus, axillae and groin. Specimens should be collected 24–48 hours after birth.
- Type-specific antibody assays are commercially available and may be useful in some clinical situations (though availability in Canada is currently limited): (a) patients with apparent first-episode genital herpes with a negative culture or NAAT; (b) identification of a seropositive pregnant woman with no history of herpes; (c) counselling HSV serologically discordant couples.¹⁸

5. *Treponema pallidum* (syphilis)

- Consult with your laboratory on tests available.
- When lesions are present in primary, secondary or early congenital syphilis, clear serous fluid should be collected for dark-field microscopy, enabling observation of morphology and movement of the spirochetes (not reliable for oral or rectal lesions).¹⁹
- Other non-serological methods involve direct fluorescent antibody tests or NAATs. The latter are very sensitive and specific.²⁰
- In cases where pregnant women are suspected of having syphilis, sections of placenta should be collected at birth and sent for DFA testing. (See *Syphilis* chapter for comprehensive recommendations on the management of syphilis in pregnant women).
- Serological diagnosis involves initial screening of sera by non-treponemal tests such as the Venereal Disease Research Laboratory (VDRL), rapid plasma reagin (RPR), toluidine red unheated serum test (TRUST) or the reagin screening test (RST).
- The introduction of treponemal tests for IgG/IgM antibodies, such as the treponemal enzyme immunoassay (EIA), may provide a more sensitive screening test for syphilis. Although EIA is highly sensitive, the test can lack specificity therefore if the treponemal-specific ELISA is positive, confirmation by a second treponemal-specific test is required (e.g. TP-PA, MHA-TP, FTA-ABS).
- Sera positive in non-treponemal tests are retested by treponemal assays such as the *Treponema pallidum* particle agglutination (TP-PA) test, fluorescent treponemal antibody absorption (FTA-ABS) test and microhemagglutination for *Treponema pallidum* (MHA-TP).²¹ Several enzyme immunoassays (EIA) have been developed commercially to detect IgG or IgM to specific *T. pallidum* antigens and are useful in HIV co-infected patients. See *Syphilis* chapter for information on cerebrospinal fluid examination.
- Treponemal tests (e.g. FTA-ABS, MHA-TP and EIA) usually remain reactive for life regardless of treatment, although 15–25% will serorevert if the patient is treated during the primary stage.

6. Human Immunodeficiency Virus

- HIV diagnostic laboratories in Canada are instructed to use only tests approved by Health Canada.
- Sera are initially screened by EIA and may detect antibodies by 3 weeks after infection, but can take up to 6 months.²²
- All positives are confirmed using a different EIA or Western blot.
- Qualitative PCR is used to detect small amounts of nucleic acid in babies born to HIV-infected mothers.
- Quantitative PCR (viral load testing) is used to monitor HIV-positive patients prior to and during antiretroviral therapy.²³
- Genotyping is used to detect the development of drug resistance in selected patients, enabling physicians to choose appropriate antiretroviral drug combinations.²⁴

7. Human papillomavirus

- Liquid-based cytology has increased the accuracy of Pap testing.
- HPV signal amplification hybrid capture assay, hc2. (Digene) and AMPLICOR PCR (Roche) are approved for use in Canada and can be performed on the same or a separate cervical sample.²⁵
- The presence of high-risk HPV in patients with atypical squamous cells of undetermined significance (ASCUS) may enable recommendation for immediate colposcopy.²⁶
- Microscopy, culture and antigen detection have no proven utility for the diagnosis of HPV infections.
- Linear array HPV genotyping (Roche) and serology are used for epidemiological purposes at the present time.
- Consult with your laboratory concerning HPV testing, as few laboratories are currently providing this service in Canada.

8. Hepatitis B virus

- Patients acutely infected with HBV will have positive EIA results for hepatitis B surface antigen (HBsAg) and/or anti-hepatitis B core (anti-HBc) IgM tests performed on sera.
- Most patients (90%) develop immunity within 6 months of infection, lose their HBsAg and have it replaced by anti-HBc IgG and anti-hepatitis B surface antibodies (anti-HBs).²⁷
- Patients chronically infected will demonstrate HBsAg persistence for 6 months or more.
- The presence of hepatitis B e antigen (HBeAg) in acutely or chronically infected individuals indicates greater infectivity for contacts and for babies born to positive mothers.²⁸ These antigens may eventually be replaced by antibodies (anti-HBe).
- Quantitative PCR assays to detect viral DNA are available to monitor response to treatment.^{29,30}

9. Hepatitis A virus

- The presence of hepatitis A virus (HAV) IgM antibodies, which may be present for 3 months, is diagnostic of acute infection.³¹

- HAV IgG antibody testing can demonstrate immunity.

10. *Trichomonas vaginalis*

- The vaginal pH is >4.5 , and the whiff test is usually negative (the withdrawn speculum does not have an abnormal odour).³²
- Because of the low sensitivity of direct microscopy, culture may be used, where available, to isolate the parasite from urethral swabs, urine sediments, prostate fluid and vaginal specimens.³³

11. *Candida albicans*

- The vaginal pH is normal (<4.5), and the whiff test is negative.³⁴
- Wet-mount preparation with 10% KOH shows budding yeast and/or branching pseudohyphae.

12. Bacterial vaginosis

- The vaginal pH is >4.5 , and the whiff test is positive.³⁵
- Gram stain demonstrates a shift in vaginal flora, with a decrease in large Gram-positive rods (lactobacilli) and an increase in small Gram-variable coccobacilli and clue cells (vaginal epithelial cells covered with numerous coccobacilli).

References

1. Chernesky MA. Laboratory services for sexually transmitted diseases: overview and recent developments. In: Holmes KK, Sparling P, Mardh PA, et al, eds. *Sexually Transmitted Diseases*. 3rd ed. New York, NY: McGraw Hill; 1999: 1281–1294.
2. Schachter J, McCormack WM, Chernesky MA, et al. Vaginal swabs are appropriate specimens for diagnosis of genital tract infection with *Chlamydia trachomatis*. *J Clin Microbiol* 2003;41:3784–3789.
3. Chernesky MA. The laboratory diagnosis of *Chlamydia trachomatis* infections. *Can J Infect Dis Med Microbiol* 2005;16:39–44.
4. Chernesky M, Jang D, Chong S, Sellors J, Mahony J. Impact of urine collection order on the ability of assays to identify *Chlamydia trachomatis* infections in men. *Sex Transm Dis* 2003;30:345–347.
5. Sellors J, Lorincz AT, Mahony JB, et al. Comparison of self-collected vaginal, vulvar and urine samples with physician-collected cervical samples for human papillomavirus testing to detect high-grade squamous intraepithelial lesions. *CMAJ* 2000;163:513–518.
6. O'Brien SF, Bell TA, Farrow JA. Use of a leukocyte esterase dipstick to detect *Chlamydia trachomatis* and *Neisseria gonorrhoeae* urethritis in asymptomatic adolescent male detainees. *Am J Public Health* 1988;78:1583–1584.
7. Hedin G, Abrahamsson G, Dahlberg E. Urethritis associated with *Chlamydia trachomatis*: comparison of leukocyte esterase dipstick test of first-voided urine and methylene blue-stained urethral smear as predictors of chlamydial infection. *APMIS* 2001;109:595–600.
8. Van Dyck E, Ieven M, Pattyn S, Van Damme L, Laga M. Detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* by enzyme immunoassay, culture, and three nucleic acid amplification tests. *J Clin Microbiol* 2001;39:1751–1756.
9. Shafer M, Moncada J, Boyer CB, Betsinger K, Flinn SD, Schachter J. Comparing first-void urine specimens, self-collected vaginal swabs, and endocervical specimens to detect *Chlamydia trachomatis* and *Neisseria gonorrhoeae* by a nucleic acid amplification test. *J Clin Microbiol* 2003;43:4395–4399.
10. Serlin M, Shafer MA, Tebb K, et al. What sexually transmitted disease screening method does the adolescent prefer? Adolescents' attitudes toward first-void urine, self-collected vaginal swab, and pelvic examination. *Arch Pediatr Adolesc Med* 2002;156:588–591.
11. Gaydos CA, Quinn TC, Willis D, et al. Performance of the APTIMA Combo 2 assay for detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* in female urine and endocervical swab specimens. *J Clin Microbiol* 2003;41:304–309.
12. Clarke LM, Sierra MF, Daidone BJ, Lopez N, Covino JM, McCormack WM. Comparison of the Syva MicroTrak enzyme immunoassay and Gen-Probe PACE 2 with cell culture for diagnosis of cervical *Chlamydia trachomatis* infection in a high-prevalence female population. *J Clin Microbiol* 1993;31:968–971.
13. Mahony JB, Chernesky MA, Bromberg K, Schachter J. Accuracy of an IgM immunoassay for the diagnosis of chlamydial infections in infants and adults. *J Clin Microbiol* 1986;24:731–735.
14. Ng LK, Martin IE. The laboratory diagnosis of *Neisseria gonorrhoeae*. *Can J Infect Dis Med Microbiol* 2005;16:15–25.
15. Whittington W, Ison C, Thompson S. Gonorrhoea. In: Morse S, ed. *Atlas of Sexually Transmitted Diseases and AIDS*. 2nd ed. London: Mosby-Wolfe; 1996: 99–117.

16. Alfa M. The laboratory diagnosis of *Haemophilus ducreyi*. *Can J Infect Dis Med Microbiol* 2005;16:31–34.
17. Singh A, Preiksaitis J, Romanowski B. The laboratory diagnosis of herpes simplex virus infections. *Can J Infect Dis Med Microbiol* 2005;16:92–98.
18. Ashley RL. Sorting out the new HSV type specific antibody tests. *Sex Transm Infect* 2001;77:232–237.
19. Ratnam S. The laboratory diagnosis of syphilis. *Can J Infect Dis Med Microbiol* 2005;16:45–51.
20. Wicher K, Horowitz HW, Wicher V. Laboratory methods of diagnosis of syphilis for the beginning of the third millennium. *Microbes Infect* 1999;1:1035–1049.
21. Stoll BJ, Lee FK, Larsen S, et al. Clinical and serologic evaluation of neonates for congenital syphilis: a continuing diagnostic dilemma. *J Infect Dis* 1993;167:1093–1099.
22. Fearon M. The laboratory diagnosis of HIV infections. *Can J Infect Dis Med Microbiol* 2005;16:26–30.
23. Phillips KA, Bayer R, Chen JL. New Centers for Disease Control and Prevention's guidelines on HIV counseling and testing for the general population and pregnant women. *J Acquir Immune Defic Syndr* 2003;32:182–191.
24. Hirsch MS, Brun-Vezinet F, Clotet B, et al. Antiretroviral drug resistance testing in adults infected with human immunodeficiency virus type 1: 2003 recommendations of an international AIDS society–USA Panel. *Clin Infect Dis* 2003;37:113–128.
25. Coutlee F, Rouleau D, Ferenczy A, Franco E. The laboratory diagnosis of genital human papillomavirus infections. *Can J Infect Dis Med Microbiol* 2005;16:83–91.
26. Wright TC Jr, Cox JT, Massad LS, Twiggs LB, Wilkinson EJ; ASCCP-Sponsored Consensus Conference. 2001 consensus guidelines for the management of women with cervical cytological abnormalities. *JAMA* 2002;287:2120–2129.
27. Krajden M, McNabb S, Petric M. The laboratory diagnosis of hepatitis B virus. *Can J Infect Dis Med Microbiol* 2005;16:65–72.
28. Okada K, Kamiyama I, Inomata M, Imai M, Miyakawa Y. e antigen and anti-e in the serum of asymptomatic carrier mothers as indicators of positive and negative transmission of hepatitis B virus to their infants. *N Engl J Med* 1976;294:746–749.
29. Chu CJ, Hussain M, Lok AS. Quantitative serum HBV DNA levels during different stages of chronic hepatitis B infection. *Hepatology* 2002;36:1408–1415.
30. Lok AS, Zoulim F, Locarnini S, et al. Monitoring drug resistance in chronic hepatitis B virus (HBV)- infected patients during lamivudine therapy: evaluation of performance of INNO-LiPA HBV DR assay. *J Clin Microbiol* 2002;40:3729–3734.
31. Chernesky MA, Gretch D, Mushahwar IK, Swenson PD, Yarbough PO. Laboratory diagnosis of hepatitis viruses. *Cumitech* 1998;Nov:18A.
32. Garber GE. The laboratory diagnosis of *Trichomonas vaginalis*. *Can J Infect Dis Med Microbiol* 2005;16:35–38.
33. Beal C, Goldsmith R, Kotby M, et al. The plastic envelope method, a simplified technique for culture diagnosis of trichomoniasis. *J Clin Microbiol* 1992;30:2265–2268.
34. Hillier S, Arko R. Vaginal infections. In: Morse S, ed. *Atlas of Sexually Transmitted Diseases and AIDS*. 2nd ed. London: Mosby-Wolfe; 1996: 149–158.
35. Money D. The laboratory diagnosis of bacterial vaginosis. *Can J Infect Dis Med Microbiol* 2005; 16:77-79.