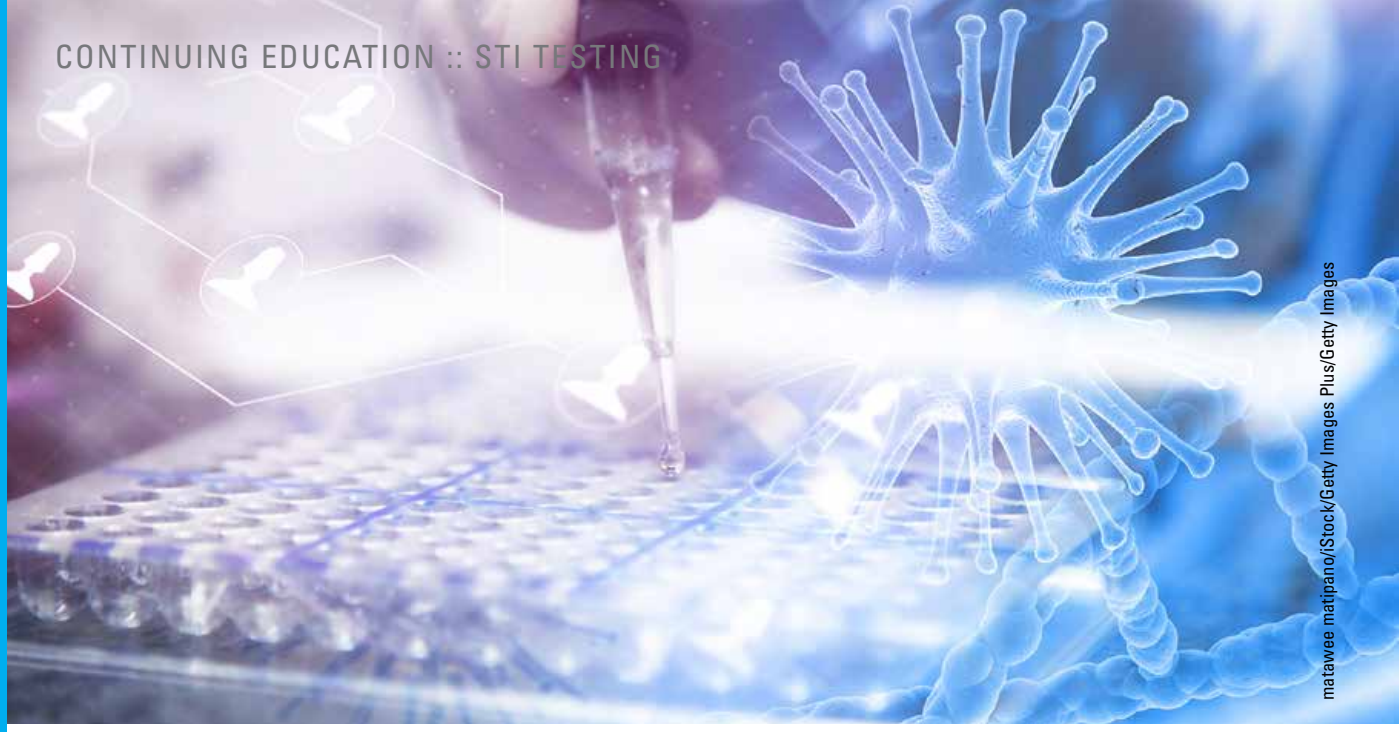

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Testing for sexually transmitted infections

By Rajasri Chandra, MS, MBA

Infections transmitted through sexual contact, including vaginal, anal, and oral sex are called sexually transmitted infections (STI)s. There are more than 30 different bacteria, viruses, and parasites that can cause STIs.[1] And some infections can be transmitted from mother to child during pregnancy, childbirth, and breastfeeding.[1] STIs are a global health problem, including in the United States. According to the Centers for Disease Control and Prevention's (CDC's) 2022 STI Surveillance Report, more than 2.5 million cases of Chlamydia, gonorrhea, and syphilis were reported in the United States, making STI a great public health concern.[9]

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LEARNING OBJECTIVES

Upon completion of this article, the reader will be able to:

1. List the main sexually transmitted pathogens and the diseases they cause.
2. Discuss STIs that can result in a risk of cancer.
3. List and describe the test methodologies used for diagnosis of STIs and the benefits of each.
4. Describe the type of settings that STI testing is used in.

Most common STIs and the diseases they cause

The most common STI-causing pathogens and the diseases they cause are provided in **Table 1** below:[2]

Emerging STIs

Some emerging outbreaks acquired by sexual contact and their diseases are listed in **Table 2** below:

Consequences of STIs[10]

STIs affect fertility

STIs, if left untreated, cause damage to the reproductive organs. STIs like Chlamydia, Neisseria gonorrhea, and Trichomatis cause infection of the uterus, fallopian tubes, and ovaries in females, resulting in inflammation, i.e., swelling and scarring of these organs leading to a condition called Pelvic inflammatory disease (PID). According to a CDC estimate, more than 1 million women are annually diagnosed with PID in the United States.[11] The inflammation and scarring from these infections can be permanent and result in ectopic pregnancy or infertility later in life.

STIs increase the risk of cancer

STIs have been shown to increase the risk of developing certain cancers. The STI infections that can cause cancer are:

Human papillomavirus (HPV) – HPV is a group of over 200 viral genotypes. They can be transmitted through vaginal, anal, or oral sex. Based on their risk of causing cancer, they are classified as high risk or oncogenic HPV and low risk or non-oncogenic type.[12] High-risk HPVs can cause several types of cancer — anal, cervical, oropharyngeal, penile, vaginal, and vulvar. [12] There are 12 high-risk HPV types: HPV 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, and 59. HPV 16 and HPV 18 are

responsible for most HPV-related cancers.[12]

Low-risk HPV types rarely cause cancer, although a few low-risk HPV types can cause warts on or around the genitals, anus, mouth, or throat. When warts form in the larynx or respiratory tract, it leads to a condition called respiratory papillomatosis, which can cause breathing problems.[12]

HPV infections are often cleared by the host's innate defense mechanism. It is believed, up to 90% of high-grade, pre-cancerous lesions in young women regress without treatment in two years. Only 5% of HPV infections progress to CIN 2 or CIN 3 within three years. Of those that progress, 80% CIN 3 lesions regress, and approximately 20% progress to invasive carcinoma within five years. Of this 20%, only 40% progress to invasive carcinoma within 30 years. HPV infection can lead to cervical cancer only if the high-risk HPV type(s) persist for long enough to cause abnormalities in the cervix.[13]

Hepatitis B virus (HBV) — HBV is transmitted through blood, semen, or another body fluid from a person infected with the virus to someone who is uninfected by sexual contact; sharing needles, syringes, or other drug-injection

Pathogen	Clinical manifestation and other associated diseases
Discharge causing infections	
Neisseria gonorrhea (NG)	**GONORRHEA** • Men: urethral discharge (urethritis), epididymitis, orchitis, infertility • Women: cervicitis, endometritis, salpingitis, pelvic inflammatory disease, infertility, preterm rupture • of membranes, perihepatitis; commonly asymptomatic • Both sexes: proctitis, pharyngitis • Neonates: conjunctivitis
Chlamydia trachomatis (CT)	**CHLAMYDIAL INFECTION** • Men: urethral discharge (nongonococcal urethritis), epididymitis, orchitis, infertility • Women: cervicitis, endometritis, salpingitis, pelvic inflammatory disease, infertility, ectopic pregnancy, preterm rupture of membranes, perihepatitis; commonly asymptomatic • Both sexes: proctitis, pharyngitis, Reiter's syndrome • Neonates: conjunctivitis, pneumonia
Trichomonas vaginalis (TV)	**TRICHOMONIASIS** • Men: urethral discharge (nongonococcal urethritis); often asymptomatic • Women: vaginosis with profuse, frothy vaginal discharge; preterm birth, low-birth-weight babies • Neonates: low birth weight
Mycoplasma genitalium	• Men: urethral discharge (nongonococcal urethritis) • Women: cervicitis, endometritis, probable pelvic inflammatory disease; often asymptomatic
Lesion and genital ulcer-causing infections	
Treponema pallidum (TP)	**SYPHILIS** • Both sexes: primary ulcer (chancre) with local adenopathy, skin rashes, condylomata lata; bone, cardiovascular and neurological damage • Women: pregnancy loss (abortion, stillbirth), premature delivery • Neonates: stillbirth, congenital syphilis
Herpes simplex virus type 2 (HSV-2) **Herpes simplex virus type 1** (HSV-1) (less commonly)	**GENITAL HERPES** • Both sexes: anogenital vesicular lesions and ulcerations • Neonates: neonatal herpes (often fatal)
Human papillomavirus (HPV)	**GENITAL WARTS** • Men: penile and anal warts; carcinoma of the penis • Women: vulval, anal and cervical warts, cervical carcinoma, vulval carcinoma, anal carcinoma • Neonates: laryngeal papilloma
Molluscum contagiosum virus	**MOLLUSCUM CONTAGIOSUM** • Both sexes: genital or generalized umbilicated, firm skin nodules
Haemophilus ducreyi	**CHANCROID** • Both sexes: painful genital ulcers; may be accompanied by bubo
Klebsiella (Calymmatobacterium) granulomatis	**DONOVANOSIS (GRANULOMA INGUINALE)** • Both sexes: nodular swellings and ulcerative lesions of the inguinal and anogenital areas
Systemic viral infections	
Human immunodeficiency virus (HIV)	**ACQUIRED IMMUNODEFICIENCY SYNDROME (AIDS)** • Both sexes: HIV-related disease, AIDS
Hepatitis B Virus (HBV)	**VIRAL HEPATITIS** • Both sexes: acute hepatitis, liver cirrhosis, liver cancer
Cytomegalovirus	**CYTOMEGALOVIRUS INFECTION** • Both sexes: subclinical or nonspecific fever, diuse lymph node swelling, liver disease, etc.
Kaposi sarcoma associated herpesvirus (human herpesvirus type 8)	**KAPOSI SARCOMA** • Both sexes: aggressive type of cancer in immunosuppressed individuals
Fungal infections	
Candida albicans	**CANDIDIASIS** • Men: superficial infection of the glans penis • Women: vulvo-vaginitis with thick curd-like vaginal discharge, vulval itching or burning
Parasitic infestations	
Phthirus pubis	**PUBIC LICE INFESTATION**
Sarcoptes scabiei	**SCABIES**

Table 1. Main sexually transmitted pathogens and the diseases they cause.

equipment; or from the gestational parent to baby during pregnancy or at birth.

For some, hepatitis B is an acute, or short-term, illness; for others, it can become a long-term, chronic infection. Chronic hepatitis B can lead to cirrhosis, liver cancer, and even death.[14]

Human immunodeficiency virus (HIV) — HIV may not cause cancer directly, but over time it causes the immune system to weaken, putting people living with HIV (PLWH) at an increased risk of developing opportunistic cancers, considered as AIDS-defining cancers. The commonly observed cancers are Kaposi sarcoma, non-Hodgkin lymphoma, and cervical cancer.[15] In addition, many STIs including Chlamydia, Neisseria gonorrhea, HSV have been shown to increase the risk for HIV infection.

Symptoms of STIs

Some of the common symptoms of STIs are as follows:[16]

- Unusual discharge from the vagina, penis, or anus
- Pain when peeing
- Lumps or skin growths around genitals or anus
- Rash
- Unusual vaginal bleeding
- Itchy genitals or anus
- Blisters, sores, or warts around genitals or anus
- Warts in the mouth or throat, but this is very rare

Asymptomatic STIs

Often STIs show no apparent symptoms. Asymptomatic rates[10] for some of the common STIs are shown below in **Table 3**:

Types of tests for STIs

Testing for STIs is always high in demand and a broad array of tests are available for detection of STIs.

Tests for the direct detection of the STI-causing microbe use the following techniques:[2]

Microscopy – A simple, rapid, and inexpensive test that can be performed at the doctor's office to help physicians get a presumptive diagnosis and recommend proper treatment to manage patients. The sensitivity is dependent on the user's skills and tends to be lower in asymptomatic patients.[17, 18] The analysis needs to be performed within 10 minutes of sample collection and so unsuitable for testing in reference laboratories.[19]

Pathogen	Diseases
Shigella sonnei	**Shigellosis,** a gastrointestinal infection, mostly transmitted in men having sex with men (MSM)[3]
Neisseria meningitides	**Urethritis** in men having sex with men[4]
Chlamydia trachomatis (serovars L1–L3)	**Lymphogranuloma venereum** (LGV)[5] • Both sexes: ulcer, inguinal swelling (bubo), proctitis
Monkey Pox Virus	**mPox** (MONKEYPOX)[6] • Both sexes: ulcerative rash at the site of intimate contact; proctitis
Ebola	**Ebola Virus Disease** (EVD)[7] • Both sexes
Zika	**Zika Virus** (ZIKV)[8] • Both sexes

Table 2. Pathogens causing emerging outbreaks acquired by sexual contact.

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Culture – Some STI-causing organisms can be grown using specific media with good sensitivity and specificity. Though it may take days, it is the only reliable method to test for antimicrobial resistance. However, some STIs such as Treponema pallidum and HPV cannot be cultured and culturing Chlamydia trachomatis and HSV require specialized laboratories making it very expensive. Neisseria gonorrhea samples are very fragile and often die before they reach the laboratory for culture, making the test less sensitive. The efficiency and precision of culture has been recently increased with the use of matrix-assisted laser desorption/ionization–time-of-flight (MALDI-TOF) mass spectrometry.[20]

Detection of antigens – Antigen detection-based assays allow the diagnosis of current infections. Commercially available antigen-based rapid detection tests (RDTs) are easy to use and require minimal skill. Some RDTs use lateral flow immunochromatographic (ICT) assays that can also be used at the point-of-care (POC) enabling doctors to diagnose and treat patients in one visit. However, they are not as sensitive as nucleic acid–based tests and some tests may also show false positive results.[20]

Detection by molecular method/nucleic acid-based test – Detection of STIs using nucleic-acid (DNA or RNA) amplification technique is now considered the gold standard for the diagnosis of many STIs. Nucleic acid amplification tests (NAATs) have a significantly lower turnaround time than culture, have a significantly higher sensitivity than all other tests, and may be used to screen asymptomatic cases. Though molecular tests need skilled personnel, many NAATs have been standardized and automated requiring minimal skills, and they can be multiplexed, allowing simultaneous detection of multiple organisms from the same sample. Some NAATs can also become POC tests for their ease of use. There are also many other nucleic acid–based tests that may or may not use amplification technique, which are also highly sensitive and specific.[20]

Detection of host response or antibodies to infection – Serological tests are useful for diagnosis, monitoring therapy,

STI causing organism	Rate of asymptomatic infections
Chlamydia	50% males and 70% females
Neisseria gonorrhea	Up to 40% males and at least 50% females
Herpes simplex virus (HSV)	~70% of males and females
Human papillomavirus (HPV)	~70 to 90% males and females

Table 3. Rate of asymptomatic infections in some STIs.

Pathogen	Method	Sample Type	Sensitivity	Specificity	Technical Complexity/Cost
NG	Microscopy (Gram staining)	Swabs (EC, UR, CO)	Low for women & asymptomatic men; high for men	Low for women & asymptomatic men; high for men	Low/low
	Culture (Selective media)	Swabs (EC, UR, CO, OP, VA, RE)	Moderate–high	Very high	Moderate/moderate
	Ag detection	Swabs (EC, CE, VA), urine	Low–moderate	High	Low–moderate/ moderate
	Molecular	Swabs (EC, UR, CO, OP, VA, RE), urine	Very high	Moderate–very high	Low for IA; high for PCR
CT	Microscopy (DFA)	Swabs (EC, UR, CO, OP, RE)	Low	High	Moderate/low
	Culture (mammalian cells)	Swabs (EC, UR, CO, OP, RE)	Moderate–high	Very high	High/moderate
	Antigen detection (OI, ICT, biosensors)	Swabs (EC, VA, UR), urine	Low–moderate	Very high	Low–moderate (both)
	NAAT (PCR, IA)	Swabs (EC, UR, CO, OP, VA, RE), urine, liquid cytology medium	Very high	Very high	Low for IA; high for PCR
TV	Microscopy (wet mount)	Swabs (VA, UR), urine (males)	Low–high for SYM women	Very high	Moderate/low
	Culture (selective media)	Swabs (VA, UR), urine (males)	Moderate–high for symptomatic women	Very high	Moderate/moderate
	Antigen detection (ICT)	Vaginal swab	High	Very high	Low/moderate
	NAAT (PCR, IA)	Swabs (EC, UR, CO, OP, VA, RE), urine	Very high	Very high	Low for IA; high for PCR
M. genitalium, M. hominis, U. urealyticum	NAAT (PCR, IA)	Swabs (EC, VA, UR), urine	Very high	Very high	Low for IA; high for PCR
HSV 1, 2	Microscopy (Tzanck/ Papanicolaou/ Romanovsky/ immunoperoxidase staining, DFA)	Skin/mucosal lesions, conjunctival/corneal smears, biopsies, base of vesicles/ vesicular fluid smears, tissue sections, swabs	Low for asymptomatic stains; moderate for DFA	Low for most stains; high for immunoperoxidase staining/DFA	Low–moderate/low
	Culture (mammalian cells)	Swab, skin lesions, fluid/exudate from vesicle base, mucosal sample without lesions, biopsies, conjunctival/ corneal smear	Low–high (depending on the clinical context)	High	High/high
	Antigen detection (ELISA, ICT)	Swab, vesicular fluid, or exudate from base of vesicle	High for fluid/ exudate; low for swabs	High	Low–moderate /moderate
	NAAT (PCR, IA)	Swab, skin lesions, fluid/ exudate from vesicle base, mucosal sample without lesions, aqueous/ vitreous humor, corticospinal fluid, blood	Very high	Very high	Moderate–high
T. pallidum subsp. pallidum	Microscopy (immunohistochemistry, DFA, DF)	Lesions, exudates	Low for DF; high for DFA	Low for DF; high for DFA	Moderate/low
	Immunoassays (ICT)	Whole blood, serum, plasma	Moderate–high	High	Low/moderate
	NAAT (PCR, IA)	Swabs, lesions, blood, cerebrospinal fluid, semen	Highly variable	High	Low for IA; high for PCR
HPV	NAAT (PCR, IA)	Swabs (EC), scrapes, tissue biopsies	Very high	Very high	Moderate–high (both)

Table 4a. Description of STI testing method, along with sensitivity, specificity, complexity.

Pathogen	Method	Throughput/ Automation	Multiplexing	Uses	Point-of-care potential
NG	Microscopy (Gram staining)	Moderate/no	No	Diagnosis	Yes
	Culture (Selective media)	Moderate/no	No	Diagnosis, AMR testing	No
	Ag detection	Moderate/no	Yes	Diagnosis, screening (potential)	Yes
	Molecular	High/possible	Yes	Diagnosis, screening, AMR testing (potential)	Yes
CT	Microscopy (DFA)	Moderate/no	No	Diagnosis (recommended for CO swabs)	No
	Culture (mammalian cells)	Low/no	No	Diagnosis, AMR testing, genotyping	No
	Antigen detection (OI, ICT, biosensors)	Low/no	Yes	Diagnosis, screening (potential)	Yes
	NAAT (PCR, IA)	High/possible	Yes	Diagnosis, screening	Yes
TV	Microscopy (wet mount)	Low/no	No	Diagnosis	Possible
	Culture (selective media)	Low/no	No	Diagnosis, AMR testing, genotyping	No
	Antigen detection (ICT)	Low/no	No	Diagnosis, screening	Yes
	NAAT (PCR, IA)	High/possible	Yes	Diagnosis, screening	Yes
M. genitalium, M. hominis, U. urealyticum	NAAT (PCR, IA)	High/possible	Yes	Diagnosis, screening	Possible
HSV 1, 2	Microscopy (Tzanck/ Papanicolaou/Romanovsky/ immunoperoxdase staining, DFA)	Moderate/low	No	Diagnosis, genotyping (only immunoperoxidase staining/DFA)	No
	Culture (mammalian cells)	Low/no	No	Diagnosis, genotyping	No
	Antigen detection (ELISA, ICT)	Low/no	No	Diagnosis, screening	Yes
	NAAT (PCR, IA)	High/possible	Yes	Diagnosis, screening, genotyping	Possible
T. pallidum subsp. pallidum	Microscopy (immunohistochemistry, DFA, DF)	Moderate/no	No	Diagnosis	Possible (only DF)
	Immunoassays (ICT)	Low/no	No	Diagnosis, screening	Yes
	NAAT (PCR, IA)	High/possible	Yes	Diagnosis, screening	Possible
HPV	NAAT (PCR, IA)	High/possible	Yes	Diagnosis, screening, genotyping	Possible

Table 4b. Description of STI testing method, along with throughput/automation and POC potential.

The purpose of testing	Test-specific considerations
• Diagnosis • Screening • Antimicrobial susceptibility testing • Surveillance • Quality Assurance • Evaluation of syndromic diagnosis	• Performance (sensitivity, specificity, positive and negative predictive value) • Specimen collection – self collected or requires transport media • Prevalence • Patient's morbidity • Reliability – validity of the laboratory and ease of use • Feasibility – operational requirements • Resources required include financial, personnel, infrastructure, etc. • Relative importance of validating this test by the lab among other priorities

Table 5. Potential factors influencing choice of tests for STIs.

or surveillance purpose.[21] The serological test is considered the gold-standard for syphilis.[22] Serological testing was also commonly used to determine past exposure to HSV.

Detection of microbial metabolites – Certain tests detect microbial metabolites that alter the pH of genital secretions and biogenic amines to analyze presence of STIs.[2]

Tables 4a and 4b list different testing methods for a few common STIs.[20]

The factors that need to be kept in mind when selecting a test are shown in **Table 5**:[2]

Conclusion

Sexually transmitted infections (STIs) pose far-reaching implications with respect to social, economic, and public health globally, including in the United States. In the United States, it has been reported that over 50 percent of infections are in the age group of 15 to 24.[23] To prevent and control STIs, a multi-pronged approach needs to be taken that includes education, early disease diagnosis, and treatment. To monitor the STI epidemic, the U.S. Department of Health and Human Services (HHS) has launched a 5-year STI Implementation Plan[24] and intends to monitor through 2030. That said, it is the responsibility of all sexually active men and women to use all precautions when indulging in sexual activity, be proactive in testing for sexual infections, and getting treated if found infected. STI testing is available in various settings including clinical laboratories, point-of-care tests in doctor's offices and hospitals, and home-based tests. The availability of point-of-care tests and home-based tests have made testing for STIs easier. With advancement in molecular technologies, we can hope to have more point-of-care tests that are accurate, rapid, and easy to use with reasonably low cost that can be used to diagnose and treat STIs early and curb the spread of STIs.

Scan code to go directly to the CE test.

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Rajasri Chandra, MS, MBA is a global marketing leader with expertise in managing upstream, downstream, strategic, tactical, traditional, and digital marketing in biotech, in vitro diagnostics, life sciences, and pharmaceutical industries. Raj is an orchestrator of go-to-market strategies driving complete product life cycle from ideation to commercialization.