The diagnostics landscape for sexually transmitted infections

World Health Organization

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Innovation in Global Health

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ISBN 978-92-4-007712-6 (electronic version)
ISBN 978-92-4-007713-3 (print version)

Suggested citation. The diagnostics landscape for sexually transmitted infections. Geneva: World Health Organization; 2023. Licence: CC BY-NC-SA 3.0 IGO.

Cataloguing-in-Publication (CIP) data. CIP data are available at http://apps.who.int/iris.

Design and layout by 400 Communications.

Contents

Acknowledgements

This report was prepared by **Debi Boeras** and **James S. Cohan** (Global Health Impact Group), in collaboration with **Lara Vojnov** and **Teodora Wi** of the World Health Organization (WHO) Department of Global HIV, Hepatitis and Sexually Transmitted Infections Programmes (HHS), **Igor Toskin** of the WHO Department for Sexual and Reproductive Health and Research, and **Kelsey Barrett** and **Anisa Ghadreshenas** (Unitaid). **Ismail Maatouk** (WHO HSS) and **Daniel McCartney** (independent consultant) supported the publication of this report.

Funding

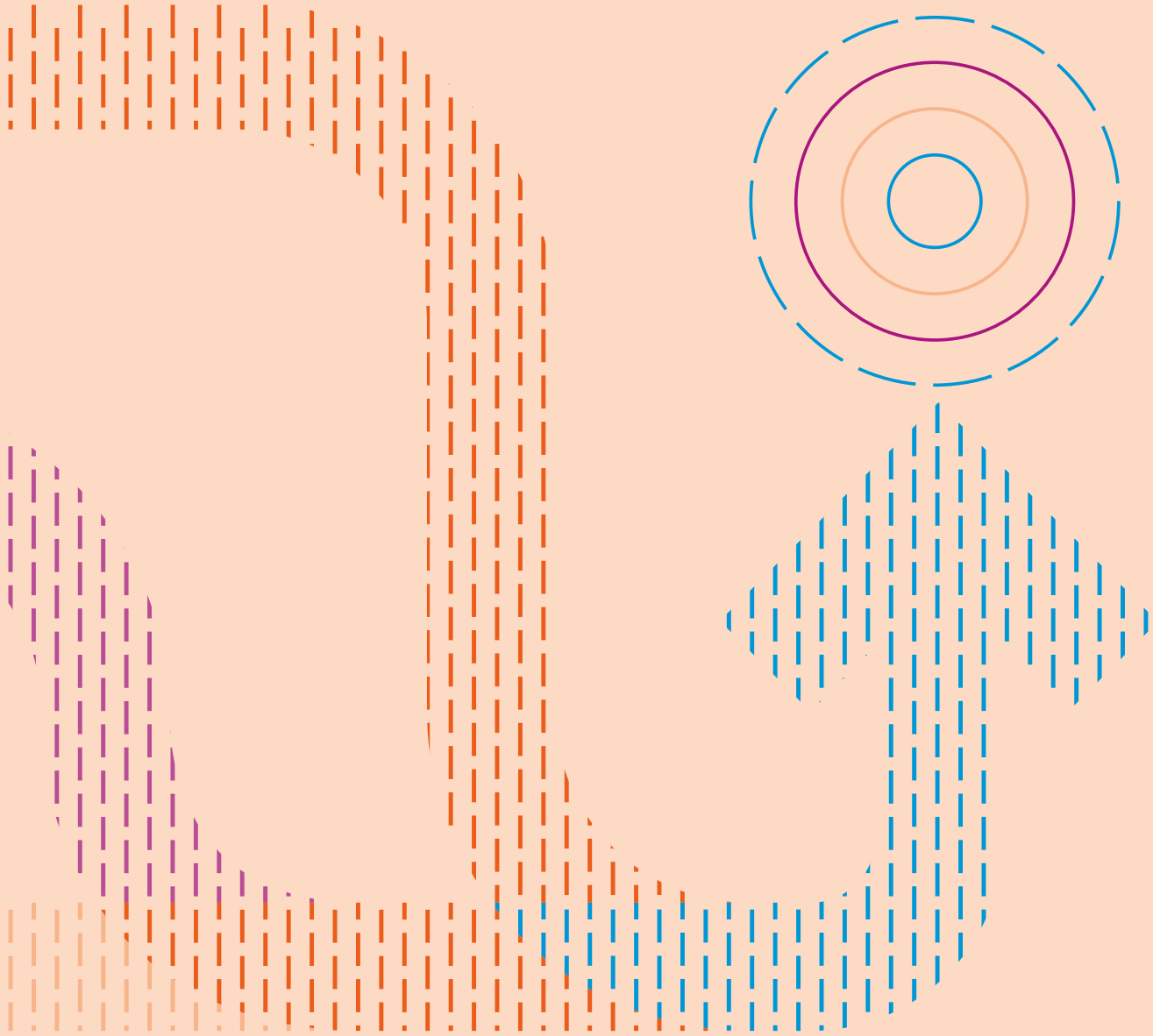
The WHO Department of the Global HIV, Hepatitis and STIs Programmes, the UNDP-UNFPA-UNICEF-WHO-World Bank Special Programme of Research, Development and Research Training in Human Reproduction (HRP) and Unitaid provided funding for this guideline.

Abbreviations

ALP	alkaline phosphatase
AMR	antimicrobial resistance
ASC-US	atypical squamous cells of undetermined significance
CDC	Centers for Disease Control and Prevention
CE	Conformité Européenne (CE)
CIN	cervical intraepithelial neoplasia
CLEIA	chemiluminescence enzyme immunoassay
CLIA	chemiluminescence immunoassay
CLIA	Clinical Laboratory Improvement Amendments (USA)
CMIA	chemiluminescent microparticle immunoassay
CPA	cross priming amplification
DFA	direct fluorescence assay
ECL	electrochemiluminescence
ECLIA	electrochemiluminescence immunoassay
EDTA	ethylenediaminetetraacetic acid
EIA	enzyme immunoassay
ELISA	enzyme-linked immunosorbent assay
FDA	U.S. Food and Drug Administration
FTA-ABS	fluorescent treponemal antibody absorbed
gG1	glycoprotein G1
gG2	glycoprotein G2
HDA	helicase-dependent amplification
HIC	high-income country
HPA	hybridization protection assay
HPV	human papillomavirus
HSV	herpes simplex virus
ICC	invasive cervical cancer
IgG	immunoglobulin G
IgM	immunoglobulin M
IVD	in vitro diagnostic
LAMP	loop-mediated isothermal amplification
LMIC	low- and middle-income countries

MFIA	multiplexed fluorometric immunoassay
MSM	men who have sex with men
MTP	microtitration plate
NAAT	nucleic acid amplification test
OD	optical density
PCR	polymerase chain reaction
POC	point-of-care
POD	peroxidase
qPCR	quantitative PCR
RDT	rapid diagnostic test
RFI	relative fluorescence intensity
RGQ	Rotor-Gene Q
RLU	relative light unit
rPCR	random PCR
RPR	rapid plasma reagin
rRNA	ribosomal RNA
RT–PCR	PCR with reverse transcription
SAC	sample adequacy control
SDA	strand displacement amplification
STAT	same-day testing and treatment
STI	sexually transmitted infection
TMA	transcription-mediated amplification
TPHA	*T. pallidum* hemagglutination
TPLA	*T. pallidum* latex agglutination
TPPA	*T. pallidum* particle agglutination
USA	United States of America
VDRL	venereal disease research laboratory

1. Introduction



1. Introduction

Sexually transmitted infections (STIs) continue to be a significant global public health issue, with an annual estimated 374 million people becoming infected with one of four curable STIs: *Treponema pallidum* (syphilis), *Chlamydia trachomatis*, *Neisseria gonorrhoeae* and *Trichomonas vaginalis (1)*. In addition, more than one in seven women is estimated to have human papillomavirus (HPV) infection *(1)*. Also of importance are herpes simplex virus (HSV) type 1 (HSV-1) and type 2 (HSV-2) and *Mycoplasma genitalium*. The World Health Organization (WHO) estimates that 3.7 billion people under the age of 50 (67%) have HSV-1 infection globally, while 491 million people aged 15–49 (13%) have HSV-2 infection globally *(1, 2)*. As to *M. genitalium*, which was first identified in the 1980s, there are little to no data with regard to its population incidence and prevalence. However, in its treatment guidelines in 2015, the Centers for Disease Control and Prevention (CDC) in the United States of America (USA) identified *M. genitalium* as an "emerging issue" *(3, 4)*. Of further concern, STIs burden low- and middle-income countries (LMICs) disproportionately; women and children are particularly vulnerable, and key populations (female sex workers, men who have sex with men) are at higher risk, understudied and underserved *(5)*. Also, scale up of interventions and integration of services are lagging.

This landscape on the diagnostics for STIs considers laboratory-based, as well as near point-of-care (POC) and true POC diagnostics for syphilis, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV (types 1 and 2) and HPV. Often the "POC" and "near POC" classifications can be ambiguous and dependent on the setting and intended use. There are now many tests available identified for use "at or near the POC" for STIs *(6)*. These include a wide range of rapid diagnostic tests as well as simple molecular tests for use in primary health care settings.

This report is intended to complement WHO's updated manual titled *Laboratory and point-of-care diagnostic testing for sexually transmitted infections, including HIV,*[1] which addresses general laboratory methods in greater detail. The focus of this landscape is on the technologies commercially available at the time of publication for syphilis, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV (types 1 and 2) and HPV. Every possible effort was made to provide a comprehensive landscape with information relevant to national programme managers. This report does not provide market assessments or pipelines for new technologies but recognizes the need for additional complementary documents to support decision-makers in prioritizing STI testing in LMICs.

The first section of this landscape provides an introduction to the different STIs that will be covered and their public health burden, with a brief overview of the types of methods that can be used to identify each. Additional details and information can be found in WHO's laboratory manual. The second section is the diagnostics landscape and includes available tests (i.e. lateral flow, molecular), platforms and systems for use in resource-limited settings where diagnostic access and delivery are challenging.

The prioritization of STIs remains low despite the urgent need for accurate screening and treatment. This report highlights the diagnostics available now to support scaled up and expanded access to screening to meet the growing testing demands in LMICs.

1.1 Methodology and scope

This report is current through 31 July 2022 using the following sources: publicly available information; published and unpublished reports and prospectuses; and interviews with developers and manufacturers. This landscape aims to provide a broad overview of key STI diagnostic assays that are commercially available. The landscape focuses on non-phenotypic methods and some of the more commonly used available methods relevant to identifying STIs in low- and middle-income countries (LMICs). Descriptions of the techniques, methods (phenotypic and non-phenotypic) and use cases are described in WHO's updated manual, *Laboratory and point-of-care diagnostic testing for sexually transmitted infections, including HIV.*[1]

1 Available at: [INSERT URL/IN PRESS]

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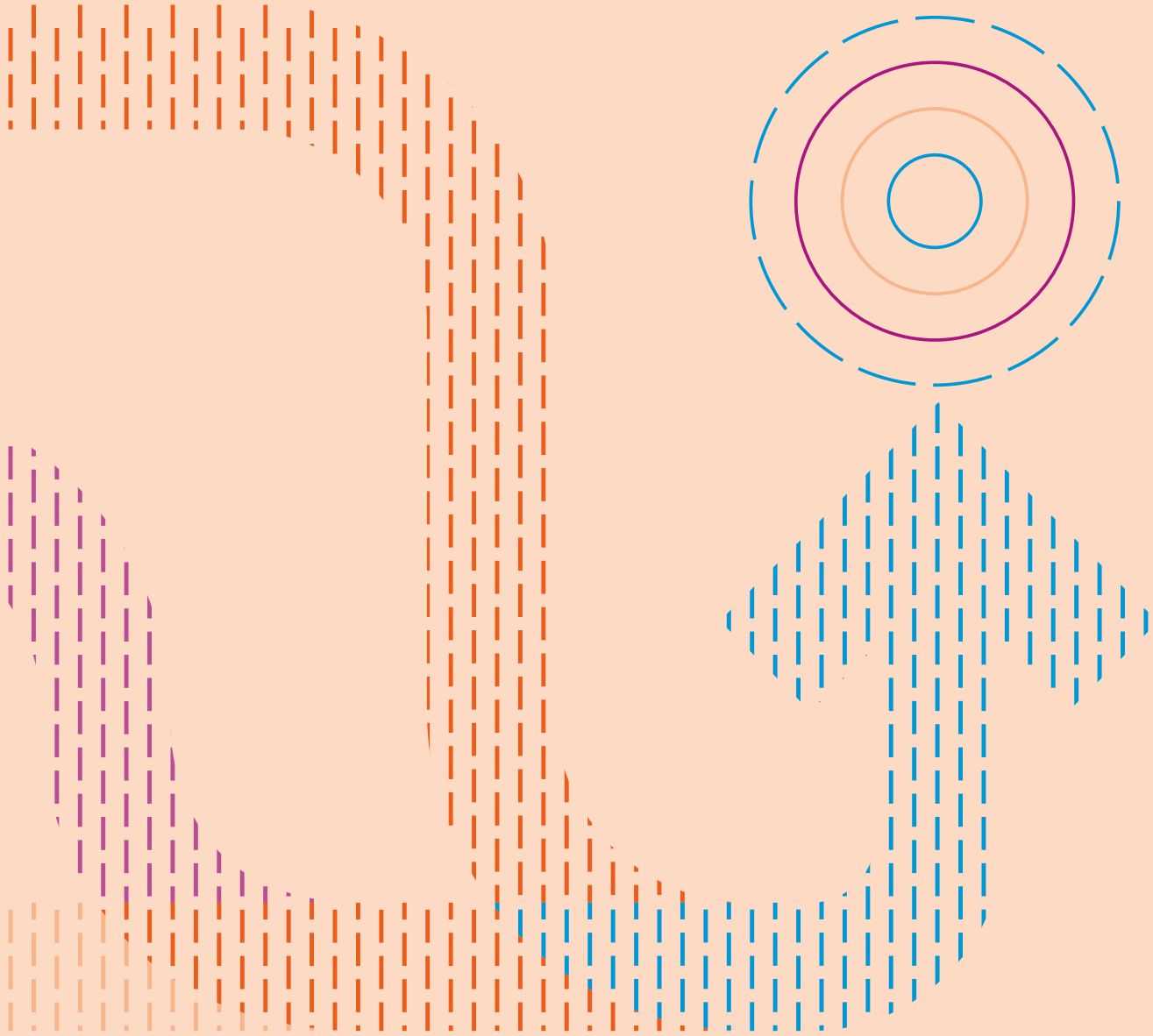
2. Global disease burden and diagnostic approaches



2. Global disease burden and diagnostic approaches

This section provides an overview of the global disease burden and magnitude of the public health problem for each STI, as well as an overview of the diagnostic approaches for each (see WHO's manual titled *Laboratory and point-of-care diagnostic testing for sexually transmitted infections, including HIV* for more details). Diagnostics play a crucial role in public health by informing clinically relevant decisions and provide quality surveillance data that can inform policy and programmes. An efficient and precise test can facilitate timely and cost-effective treatment linkage to reduce transmission.

2.1 Syphilis

The World Health Organization (WHO) estimates that in 2020, the most recent year for which such statistics are available, there were approximately 7 million new cases of syphilis worldwide *(1)*. The highest disease burden for syphilis is in the African Region *(7, 8)*. Syphilis is caused by the helically coiled bacterium *Treponema pallidum*, which first clinically manifests as a genital skin lesion and is typically painless.

Timely treatment is cost effective and curative. Without treatment, syphilis progresses from a primary, early-stage infection to secondary, latent and tertiary stages. Primary syphilis is characterized by the appearance of lesions on the genitals (with high bacterial load), which may resolve spontaneously. Thus, patients often do not seek care during the primary stage, but rather during the secondary stage, when a rash may appear on the shoulders, arms, chest or back. After a few years, symptoms may subside and patients enter a latent phase that can last for many years. In some stages, patients may be asymptomatic, making it difficult to diagnose very early syphilis, neurosyphilis and tertiary syphilis *(4)*.

Syphilis has particularly profound consequences for pregnant women, considered to be a vulnerable population, and for individuals in certain key populations where its prevalence is high: men who have sex with men (MSM) and sex workers. With regard to pregnant women, congenital syphilis, where the mother's syphilis infection is passed to her newborn, can cause low birth weight, anaemia and jaundice, among other systemic problems; ultimately, it is a significant cause of infant mortality. In 2016, researchers estimated that approximately 661 000 cases of syphilis occurred worldwide among pregnant women, many of whom were either untreated or inadequately treated *(9)*. It was estimated that about 355 000 adverse pregnancy outcomes, including 143 000 early fetal deaths and stillbirths, 61 000 neonatal deaths, 41 000 preterm or low-weight births, and 109 000 infected infants resulted *(5)*. Although these results are an improvement over the 2012 estimates, without universal testing and treatment of syphilis in pregnancy, as many as 50% of pregnancies in women with syphilis will result in adverse outcomes, including perinatal death, prematurity and low birth weight *(10)*.

With regard to MSM, WHO estimated that syphilis infected an average of 11.8% (range 5.2% to 19.6%) of MSM in 11 of 25 reporting countries, 10% or more in seven countries in 2019 *(11)*. A recent systematic review and meta-analysis of the global prevalence of syphilis among MSM found that the global pooled prevalence from 2000 to 2020 was 7.5% compared to 0.5% among men in the general population *(12)*. In the USA, the CDC estimated that in 2017, 79.6% of syphilis cases were among MSM and the numbers are increasing *(13)*. Untreated, syphilis can lead not only to serious complications, but it also increases the risk of acquiring and transmitting HIV. This can also be said of all STIs.

Finally, according to WHO, syphilis infected more than 5% of sex workers in 11 of 32 countries and more than 10% in four countries in 2019 *(14)*. Sex workers include female, male and transgender individuals who receive money or goods in exchange for sexual services; in many places, they are very vulnerable to HIV and other STIs *(14)*.

In most high-income countries (HICs), syphilis is usually diagnosed using laboratory-based tests. Testing volume is primarily driven by routine, sometimes annual, screening of high-risk populations such as MSM and pregnant women. Because syphilis cannot generally be easily cultured, testing can include direct detection via darkfield microscopy as well as serological tests, which detect either immunoglobulin G (IgG) or immunoglobulin M (IgM) antibodies against *T. pallidum* ("treponemal tests") or antibodies against antigens (primarily cardiolipin) that are produced by the host in response to a syphilis infection, that is, "non-treponemal" tests. Darkfield microscopy requires lesion exudate or tissue.

Proper specimen collection and handling is critical both for safety reasons, as lesions are highly infectious, and to achieve the best test result in terms of specificity. While darkfield examination of *T. pallidum* can be highly specific, it requires a special microscope and the technical expertise of a trained microscopist, which may be limited in LMICs (4, 15). This landscape focuses on serological non- treponemal and treponemal tests used to screen and confirm the diagnosis of syphilis.

2.2 *Chlamydia trachomatis* and *Neisseria gonorrhoeae*

Both *C. trachomatis* and *N. gonorrhoeae* are significant global health problems. WHO estimates that approximately 128 million and 82 million new cases of *C. trachomatis* and *N. gonorrhoeae*, respectively, are diagnosed annually worldwide *(1)*. In addition, antimicrobial resistance (AMR) in *N. gonorrhoeae* is particularly problematic. With resistance to both cephalosporins, including third-generation extended-spectrum cephalosporins, and fluoroquinolones, *N. gonorrhoeae* is a multidrug-resistant pathogen. Resistance is outpacing new antibiotics for *N. gonorrhoeae*. Thus, WHO considers *N. gonorrhoeae* to be a priority microorganism for AMR monitoring in the Global Antimicrobial Surveillance System and for drug development in the context of AMR *(16, 17)*.

There are many test options that can be used to detect *C. trachomatis* and *N. gonorrhoeae*, including both culture and non-culture methods. Despite low sensitivity, culture for the detection of *C. trachomatis* and *N. gonorrhoeae* was long considered to be the "gold standard" against which other tests were compared *(18, 19)*. Non-culture methods, including enzyme immunoassay (EIAs) and direct fluorescence assay (DFAs), are also available. However, the development of Nucleic acid amplification tests (NAATs) has permanently changed the testing landscape for the detection of *C. trachomatis*, *N. gonorrhoeae*, *Trichomonas vaginalis* and other STIs. Having demonstrated strong sensitivity and specificity, NAAT testing for *C. trachomatis* and *N. gonorrhoeae* is now considered the gold standard and is currently recommended for routine use *(18)*. Available testing methods for *C. trachomatis*, *N. gonorrhoeae* and *C. trachomatis*/*N. gonorrhoeae*, both laboratory-based and point-of-care (POC) or near-POC methods, are described herein.

2.3 *Trichomonas vaginalis*

Like *C. trachomatis*, *N. gonorrhoeae* and *T. pallidum*, *T. vaginalis* is a significant health problem globally. WHO estimates that there are about 156 million new cases of *T. vaginalis* annually worldwide *(1)*. While there are limited data on the burden, *T. vaginalis* has been linked to cervical cancer in women *(20)*, impaired sperm quality in men *(21)*, as well as increased risk of acquisition and transmission of HIV and other STIs *(22, 23)*. *T. vaginalis* is associated with adverse birth outcomes *(22)*.

Diagnosis of *T. vaginalis* infection in women has traditionally been performed by microscopy of vaginal secretions, but this technique requires immediate evaluation of a wet preparation and is only about 50% sensitive compared with culture or NAAT *(24, 25)*. Diagnosis of *T. vaginalis* in men is typically from wet mount with microscopic visualization of the parasites on slide preparations from urethral or prostatic secretions *(26)*. Examination of the wet mount must be performed before the microorganisms lose motility, generally within 10 to 20 minutes after sample collection *(15)*. According to the CDC, the sensitivity of wet mount examinations is low, ranging from 51% to 65% in vaginal specimens and still lower in male specimens *(3)*.

There are several reliable laboratory-based molecular systems for *T. vaginalis* testing that can accommodate swab and urine sample type, allowing for testing that is convenient, accurate and more sensitive than traditional methods. In addition, there are also commercially available POC and near-POC platforms and combination assays that include *T. vaginalis*.

2.4 *Mycoplasma genitalium*

M. genitalium was first identified in the 1980s; since then, it has been recognized as a significant cause of male urethritis *(3)*. Although the pathogenic role of *M. genitalium* is less certain in women than men, it can be found in the vagina, cervix and endometrium. *M. genitalium* infections in women may cause cervicitis and pelvic inflammatory disease, although they are often asymptomatic *(3)*.

There are little to no data regarding population incidence and prevalence of *M. genitalium* globally. Although the CDC called *M. genitalium* an "emerging issue" in its 2015 Guidelines for STIs *(3)*, it is not an STI infection that is widely researched. In its 2021 Guidelines for STIs, the CDC indicated that *M. genitalium* is associated with symptoms of urethritis and urethral inflammation, accounting for about 10–25% of nongonococcal urethritis *(4)*. *M. genitalium*, which may be asymptomatic in both men and women, is also associated with cervicitis, pelvic inflammatory disease and infertility among women *(27)*. Recent estimates indicate that *M. genitalium* may affect more than 15% of men and women in some high-risk populations and that its prevalence is increasing *(27)*. In addition, the data also indicate that as many as 50% of women and 42% of men with *M. genitalium* may have an antibiotic-resistant strain *(27)*. In particular, some strains of *M. genitalium* are resistant to macrolides (e.g. azithromycin).

Regarding diagnostic considerations, *M. genitalium* is a fastidious obligate intracellular bacterium and is extremely difficult and time-consuming to culture, taking weeks to months to achieve a result. This greatly limited testing for *M. genitalium* until the introduction of

molecular methods, which are now the gold standard for detecting *M. genitalium*. Both polymerase chain reaction (PCR)-based and transcription-mediated amplification (TMA)-based molecular tests are commercially available for *M. genitalium*. In addition, POC and near-POC platforms are available to detect *M. genitalium*. Simpler and quicker diagnostic testing enables the collection of a greater amount of data, which aids in informing programmes and identifying research needs.

In addition, because macrolide-resistant strains of *M. genitalium* have emerged and are increasing, it is recommended that all *M. genitalium*-positive assays be reflexed to a test for resistance *(28)*. Until recently, these tests could be performed primarily by sequencing methods, which are not generally available in LMICs. However, currently, molecular assays for the detection of resistance in *M. genitalium* are commercially available.

2.5 Herpes simplex virus types 1 and 2

There are two types of herpes simplex virus (HSV), HSV-1 and HSV-2. Both are chronic viral infections that are lifelong. Either type of HSV can cause genital herpes. Although HSV-1 is primarily transmitted via oral-to-oral contact causing oral herpes (e.g. mouth sores), it can also cause genital herpes, which is occurring particularly in young women and MSM *(4)*. However, HSV-2, which is transmitted by direct sexual contact, causes the most recurrent cases of genital herpes *(4)*.

HSV is a significant issue worldwide. WHO estimates that 3.7 billion people under the age of 50 (67%) have HSV-1 infection globally, while approximately 491 million people aged 15–49 (13%) have HSV-2 infection globally *(29)*.

Although HSV infections can cause painful blisters or ulcers at the site of infection, many such infections are asymptomatic, which complicates its diagnosis (3, 29). Virological testing of samples from active genital herpes lesions is the best way to diagnose the infection *(30)*. While viral cell culture has been the gold standard, more recently, molecular testing has been shown to be more sensitive.

HSV infections should be differentiated using type-specific serological tests to determine which type of HSV is causing the infection *(3)*. However, because "the first episode of symptoms of genital HSV-1 infection cannot be clinically differentiated from genital HSV-2 infection", initial treatment is agnostic regardless of HSV type *(31)*.

2.6 Human papillomavirus

Human papillomavirus (HPV) is of global concern, with more than one in seven women estimated to have HPV infection, which is a major cause of high-grade cervical intraepithelial neoplasia (CIN) (grade 2 or higher) *(1)*. In 2018, approximately 570 000 new cases of HPV-related cervical cancer were reported; in 2019, approximately 300 000 individuals died of HPV-related cervical cancer *(1)*. The CDC stated that "HPV is so common that almost every person who is sexually active will get HPV at some time in their life if they don't get the HPV vaccine" *(32)*.

In the USA and in some countries, the Papanicolaou (Pap) test has been the gold standard for detecting cervical cancer in women over 30 years of age, most of which is caused by HPV. WHO and the U.S. Food and Drug Administration (FDA) recommend that first-line screening be performed using molecular-based testing.[2] Unfortunately, most molecular-based testing is difficult in resource-limited settings, requiring centralized laboratory facilities using sophisticated instrumentation. There are now several POC or near-POC platform and assay options for use in resource-limited settings.

2.7 Antimicrobial resistance

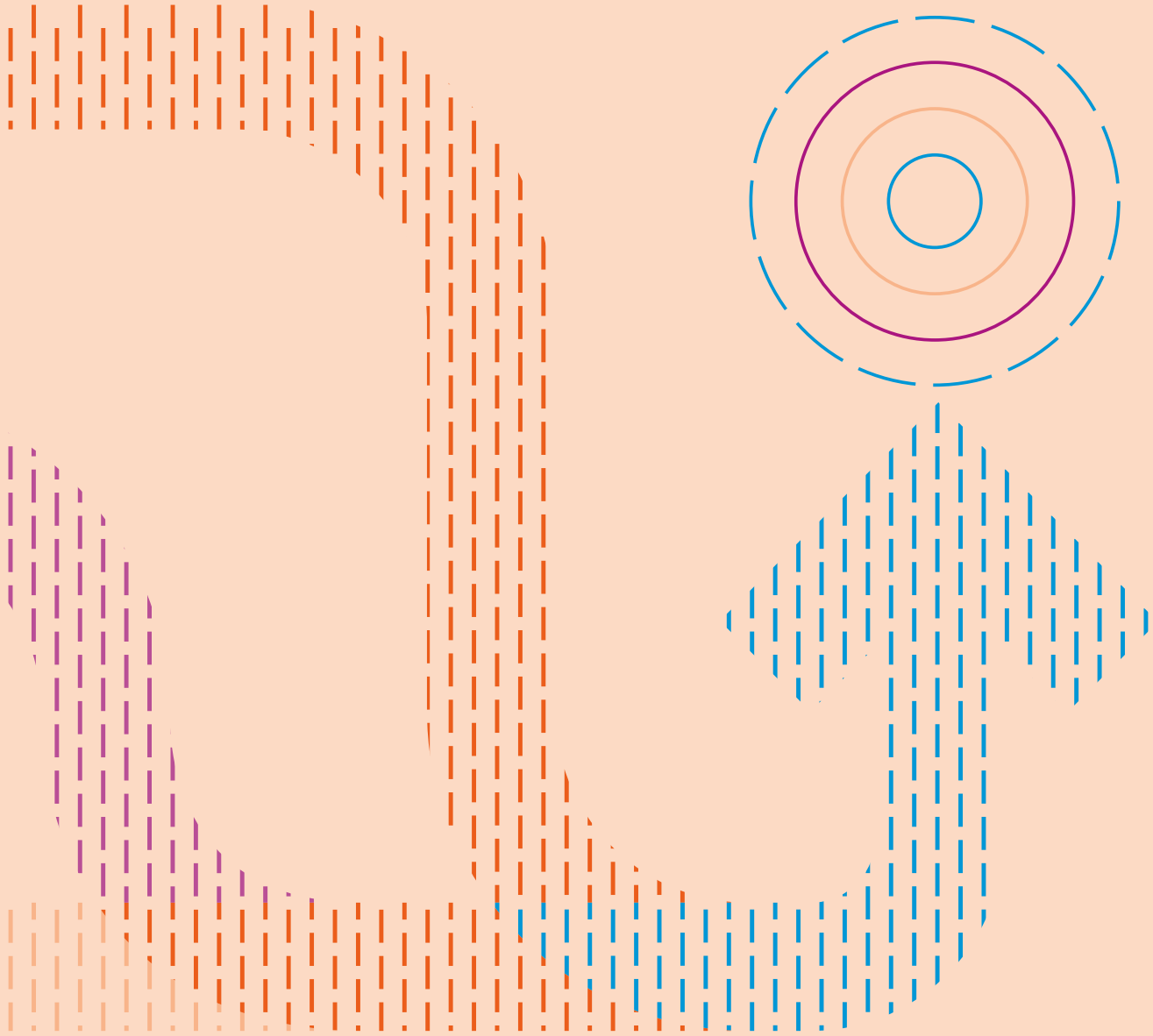
Drug resistance associated with STIs is contributing to the global spread of drug-resistant microorganisms *(33)*. A major driver for AMR in STIs is the lack of accessible diagnostics to provide clinicians with timely results. This is particularly problematic in LMICs. This manual does not focus on AMR diagnostics. This is covered in another landscape *(34)*.

2.8 In vitro diagnostics to identify STIs

There are several methods that can be used to identify each of *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HPV and HSV-1 and HSV-2. Historically, phenotypic methods of bacterial and viral identification have been the backbone of diagnostics for STIs; however, their use has many drawbacks that affect LMICs. The test methods are slow and manually cumbersome. The growth of bacteria or viruses in culture, for example, can take days (or weeks) to obtain results and there are operational challenges that include, but are not limited to, inconsistent electricity, dust, lack of climate control and human resource constraints *(35, 36)*. More recent advancements in non-phenotypic identification methods, including immunological and molecular methods, can help overcome these drawbacks and have become increasingly available and used in LMICs. Originally, molecular tests focused on detecting viruses and have since paved the way for their use in detecting bacterial pathogens *(37)*. Most molecular tests require large commercial in vitro diagnostic (IVD) platforms to be performed in high-throughput clinical laboratories; more recently, they are becoming available in a format that can be used at or near-POC.

2 Currently, in the USA, using the Pap and HPV tests in tandem (i.e. co-testing) is the preferred screening method in women over 30.

3. Diagnostics landscape



3. Diagnostics landscape

The diagnostics landscape includes available tests, platforms and systems for use in resource-limited settings where diagnostic access and delivery are challenging. The first two sections focus on syphilis as more tests are currently available due to the critical need for interventions to eliminate mother-to-child transmission of HIV and syphilis. As most are available in some combination on multiplex platforms, the final section groups all the testing methods together for the detection of *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV-1 and HSV-2, and HPV.

3.1 Tests for detecting syphilis

Syphilis testing can include serological tests, which detect either IgG or IgM antibodies against *T. pallidum* ("treponemal tests") or antibodies against antigens (primarily cardiolipin) that are produced by the host in response to a syphilis infection, that is, "non-treponemal" tests. Serological non-treponemal and treponemal tests are used to screen and confirm the diagnosis of syphilis.

Non-treponemal tests

All non-treponemal tests detect both IgM and IgG antibodies, which are commonly detectable as early as 6 days after infection with syphilis. Non-treponemal tests are useful in identifying recent syphilis infection, in monitoring the progression of syphilis and gauging the patient's response to antibiotic therapy. Non-treponemal tests typically remain reactive without treatment and can be persistently reactive for months to years after treatment. Results can be qualitative (reactive or non-reactive) and quantitative (titre); non-treponemal tests can be quantitated by serially diluting the serum until the test is no longer reactive. Non-treponemal test titres correlate with, and can be used as, a marker of disease activity and response to treatment. Decline in rapid plasma egain (RPR) titre in penicillin-treated patients follows a similar decline in the natural course of syphilis *(38)*.

The two most widely used non-treponemal serological tests are the RPR *(39)* and the venereal disease research laboratory (VDRL) tests. These tests can be run manually (visualization) or are automated. RPR detects antibodies against cardiolipin-cholesterol-lecithin egain antigens. Plasma from blood is collected and mixed with a solution of antigens along with a visualization agent (i.e. dyes, colourants). If antibodies are present, macroscopic deposits of the dyes can be seen visually. VDRLs are similar to RPR, in that they detect the presence of anti-cardiolipin antibodies; however, VDRL reactivity is visualized through foaming in a test-tube ("flocculation"), rather than with colourants as in RPR. Results for non-treponemal tests are "reactive", "weak reactive" and "non-reactive". "Positivity" of results is reserved for treponemal tests.

Commercial non-treponemal tests

Table 1 summarizes the serological non-treponemal tests commercially available. Tests are listed alphabetically according to manufacturer. If the test uses a separate platform, this has also been identified.

Table 1: Non-treponemal syphilis tests

Manufacturer/ test (platform)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
ASI Evolution Automated RPR Syphilis Test (ASI Evolution) 	Qualitative	190 per hour	N/A	Serum, plasma	Reagin antibodies	FDA CE-IVD	Automated, flocculation, integrated digital particle analyser
BD Macro-Vue RPR Card Tests	Qualitative	Manual, low-throughput	N/A	Venous blood, large serum volume	Antibodies (unspecified)	CE-IVD	Manual, flocculation
Bio-Rad BioPlex 2200 Syphilis Total & RPR 		100 samples per hour, 22 results each	20–45 min	Serum, heparinized plasma, EDTA plasma	Reagin antibodies, total IgG/IgM	FDA CE-IVD	Automated, dual treponemal/ non-treponemal MFIA, proprietary software
Gold Standard Diagnostics AIX1000 RPR Test 	Qualitative	Fits 192 samples	N/A	Serum	Reagin antibodies	FDA CE-IVD	Automated, flocculation, camera and proprietary software algorithm
SEKURE RPR and TPLA assays (SK500 Platform) 	Semi-quantitative	580 tests per hour and 72 patient sample on-board capability	10 min	Serum, plasma	Antibodies (unspecified)	FDA CE-IVD	Automated, measures light absorbance based on agglutination

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; EDTA: ethylenediaminetetraacetic acid; FDA: U.S. Food and Drug Administration; IgG: immunoglobulin G; IgM: immunoglobulin M; MFIA: multiplexed flow immunoassay; N/A: not applicable; TPLA: *Treponema pallidum* latex agglutination.

ASI Evolution Automated RPR Syphilis Test (Arlington Scientific): The ASI Evolution Automated RPR Test for syphilis for use on the ASI Evolution is a qualitative non-treponemal flocculation test for the detection of egain antibodies in human serum and plasma as a screening test for serological evidence of syphilis. The test format is a 48-well plastic plate. The test runs on the ASI Evolution platform, which can process and analyse up to 190 samples per hour. One technician can run three ASI Evolutions simultaneously, with the ability to perform over 4000 tests in an 8-hour shift.

The ASI Evolution, is an integrated digital particle analyser designed to objectively interpret certain agglutination tests by using a camera to create a highly sensitive and high-resolution image of the agglutination immunoassay. The ASI Evolution fully automates the sample and reagent handling steps of the test procedure. Sample volume is 110 µl (either serum or plasma).

BD Macro-Vue RPR Card Tests (Becton Dickinson): The Becton Dickinson (BD) Macro-Vue RPR Teardrop Card Test (using fingerstick blood) was the original Card Test developed by the manufacturer. It was developed for field use where testing could be performed without laboratory equipment. Subsequently, by incorporating machine rotation, ringed test surfaces, and by making other technical changes, BD developed the RPR 18-mm circle card test for use in large-scale testing in public health and clinical laboratories. The circle card test, which is CE-IVD-marked, is recommended when venous blood collection is used and a large volume of serum is available, which is generally the case in public health and clinical laboratories. When a specimen contains antibody, flocculation occurs with co-agglutination of the carbon particles of the RPR card antigen, which

appears as black clumps against the white background of the plastic-coated card. By contrast, nonreactive specimens have a light-grey colour.

Bio-Rad BioPlex 2200 Syphilis Total & RPR (Bio-Rad Laboratories): The BioPlex 2200 Syphilis Total & RPR assay is a multiplex flow immunoassay (MFIA) intended for the simultaneous detection of non-treponemal reagin antibodies and total IgG/IgM treponemal antibodies in human serum or plasma. It is the only dual treponemal/non-treponemal immunoassay test that offers an automated one-step testing solution for syphilis testing. The fully automated assay uses antigen-coated fluoromagnetic beads with unique fluorescent signatures to identify the presence of IgG or IgM antibodies to reagin antigens or *T. pallidum*.

The BioPlex 2200 Syphilis Total & RPR assays are performed on the BioPlex 2200 multiplexing platform. The system is a fully automated, random-access multiplex testing platform that combines automation with proprietary multiplex chemistry and the eFlex software. It is a high-throughput platform that can run 22 results per patient sample at 100 samples per hour, yielding up to 2200 results per hour. Time-to-first-result is 20–45 minutes depending on the assay panel being run.

Gold Standard Diagnostics AIX1000 RPR Automated Test (Gold Standard Diagnostics): The Gold Standard Diagnostics AIX1000 RPR Automated Test system is a non-treponemal flocculation test that can qualitatively determine the presence of reagin antibodies in human serum. It may be used to aid in the diagnosis of syphilis when used in conjunction with supplemental treponemal laboratory tests and other clinical information. This test may also be used to detect non- treponemal antibodies in samples serially diluted to establish titre information. The system consists of the AIX1000 Analyzer and RPR test reagents.

The AIX1000 Analyzer, which is CE-marked and U.S. FDA-cleared, is a fully automated system. Reagents and up to 192 patient samples can be loaded on the system. The AIX1000 automates the processing, analysis, reporting and archival of results for both RPR screens and titres. The system delivers serum from collection tubes into test wells. After the antigen suspension is added, the test wells are incubated while being shaken. An on-board camera is used to obtain a high-resolution image. This image is analysed using the proprietary software algorithm to interpret the results.

SEKURE RPR and TPLA assays (Sekisui Diagnostics): Sekisui Diagnostics manufacture the SEKURE RPR and *Treponema pallidum* latex agglutination (TPLA) assays that run on their chemistry platform SK500. The results are semi-quantitative for monitoring therapy. In its TPLA assay, polystyrene latex beads coated with antigens from *T. pallidum* bind to antibodies in the sample; agglutination of the sample results in a change in light absorbance. In the RPR assay, specific lipid antigens (cardiolipin and lecithin) coat polystyrene beads; like the TPLA assay, this results in agglutination of beads if antibodies are present.

The SK500 instrument has a throughput of up to 580 tests per hour and 72 patient sample on-board capability. The system has 36 on-board assays to meet the testing needs of small to moderate volume laboratories. The company emphasizes that the SK500 has a simple and intuitive interface that requires minimal training; use of primary tubes and cups allows for easy deployment. The turnaround time of the RPR and TPLA assays is approximately 10 minutes, excluding sample preparation.

Treponemal tests

Most treponemal tests are immunoassays that detect antibodies against treponemal antigens. They are characterized by high sensitivity and specificity, particularly for secondary syphilis, and are used mainly as confirmatory tests to verify reactivity in non-treponemal tests. Because treponemal tests can be positive for years with or without treatment, they are not recommended to evaluate response to therapy, relapse or reinfection. Also, most treponemal tests do not differentiate venereal syphilis from endemic syphilis (yaws and pinta). Types of treponemal tests include the following.

Fluorescent treponemal antibody (FTA) absorbed (FTA-ABS) test: This test is an indirect immunofluorescence staining method that uses fluorescently labelled immunoglobulins that bind to the patient's antibodies.

T. pallidum particle agglutination (TPPA) and *T. pallidum* hemagglutination (TPHA, also known as microhemagglutination assay) assays: These tests are indirect agglutination tests where antigens extracted from *T. pallidum* are coated to cells (TPHA) or gelatin particles (TPPA) and react with the patient's serum.

EIA: This test uses enzyme-labelled antigens (specific to *T. pallidum*) to detect antibodies against *T. pallidum*. The enzymes convert a substrate to a reaction product, which emits a photon of light (chemiluminescence) or develops a particular colour. Variations of the EIA include competitive, capture and sandwich methods. EIA enables higher volumes of testing and can be automated.

Molecular: Nucleic acid detection methods have been developed for the detection of *T. pallidum* in clinical specimens *(40)*, but they are not standardized in terms of target sequence, amplification methods or sample type. Several studies demonstrated sensitivities ranging from 75% to 95% depending on sample and stage. Because of the challenges with treponemal and non-treponemal tests, these molecular tests are a test of choice for congenital syphilis, neurosyphilis and early primary syphilis when traditional tests have limited sensitivity. However, there are currently no commercially available molecular assays available for the detection *T. pallidum (40)*.

Commercial treponemal tests

There are many manufacturers of treponemal assays. Some of the assays are designed for manual use, while others have been automated and approved for use on selected immunoassay systems made by other manufacturers; some are designed for use on automated systems made by the same manufacturer as the assay(s). Assays and systems for the detection of *T. pallidum* are described in **Table 2**.

Table 2: Automated treponemal systems (integrated assays and platforms)

Manufacturer/test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Abbott ARCHITECT Syphilis TP assay	Qualitative	Varies per device	Varies per device	Serum, plasma, whole blood	IgG and IgM antibodies	FDA	CMIA
Abbott ARCHITECT *i*1000$_{SR}$ 	Qualitative	65 samples at once, 100 tests per hour; low-to-medium-volume	<20 minutes	Serum, plasma, whole blood, urine	IgG and IgM antibodies	FDA	CMIA
Abbott ARCHITECT *i*2000$_{SR}$ 	Qualitative	135 samples at once, 200 tests per hour (up to 12 500 tests); medium volume	28 minutes (first results)	Serum, plasma, whole blood, urine	IgG and IgM antibodies	FDA	CMIA
Abbott ARCHITECT *i*4000$_{SR}$ 	Qualitative	Load up 285 (35 priority, 250 routine), 400 per hour; high volume	N/A	Serum, plasma, whole blood, urine	IgG and IgM antibodies	FDA	CMIA
Bio-Rad BioPlex Syphilis IgG (BioPlex 2200)	Qualitative	Multiplex	N/A	Serum	IgG antibodies	FDA	MFIA
DiaSorin LIAISON Treponema Assay	Qualitative	Varies according to the device	Varies according to the device	Serum	Total antibodies (IgG/IgM)	FDA CE-IVD	CLIA
DiaSorin LIAISON Immunotherapy Analyzer 	Qualitative	180 per hour	17 minutes (6-hour walkaway time)	Serum, plasma, urine, cerebrospinal fluid	Total antibodies (IgG/IgM)	FDA CE-IVD	CLIA, photo-multiplier
DiaSorin LIAISON XL 	Qualitative	180 per hour	17 minutes	Serum, plasma, urine, cerebrospinal fluid	Total antibodies (IgG/IgM)	FDA CE-IVD	CLIA, photo-multiplier

Table 2 (continued): Automated treponemal systems (integrated assays and platforms)

Manufacturer/test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
DiaSorin LIAISON XS 	Qualitative	85 per hour	17 minutes	Serum, plasma, urine, cerebrospinal fluid	Total antibodies (IgG/IgM)	CE-IVD	CLIA, photo-multiplier
Fujirebio Lumipulse G TP-N (G1200) 	Qualitative	120 per hour	30 minutes	Serum, EDTA plasma, citrate plasma	IgG and IgM antibodies	FDA CE-IVD	CLEIA, photometric detector
Roche Elecsys Syphilis assay	Qualitative	Varies per device	18-minute assay time	Serum, plasma	Total antibodies	Varies per device	ECLIA
Roche cobas e411 (modular part of cobas e4000 series) 	Quantitative and qualitative	86 per hour; up to 18 assays	9 minutes (first results); continuous random access	Serum, plasma, urine	Total antibodies	FDA CE-IVD	ECL
Roche cobas e601 (modular part of cobas e6000 series) 	Quantitative and qualitative	170 tests; up to 25 assays	9 minutes (first results); continuous random access	Serum, plasma, urine	Total antibodies	FDA CE-IVD	ECL
Roche cobas e602 (modular part of cobas e8000 series) 	Quantitative and qualitative	170 per hour; up to 25 assays; medium volume	Also allows for same-day testing and treatment first results	Serum, plasma, urine	Total antibodies	FDA CE-IVD	ECL
Roche cobas e801 	Quantitative and qualitative	300 per hour, per module; more than 1000 immunoassays; high volume	Continuous loading	Serum, plasma, urine, cerebrospinal fluid, supernatant, whole blood	Total antibodies	FDA	ECL
Roche cobas e4000 series	Quantitative and qualitative	Up to 530 tests per hour; low throughput	9 minutes (first results); continuous random access	Serum, plasma, urine	Total antibodies	FDA CE-IVD	ECL
Roche cobas e6000 series	Quantitative and qualitative	Loading capacity is 150 samples; medium throughput	9 minutes (first results); continuous random access	Serum, plasma, urine	Total antibodies	FDA CE-IVD	ECL

Table 2 (continued): Automated treponemal systems (integrated assays and platforms)

Manufacturer/test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Roche cobas e8000 series	Quantitative and qualitative	9800 test per hour; 440 configurations; high throughput	Also allows for same-day testing and treatment first results	Serum, plasma, urine	Total antibodies	FDA CE-IVD	ECL
Siemens ADVIA Centaur Syphilis assay (XP and CP)  	Qualitative	180 per hour (CP), 240 per hour (XP)	15 minutes (CP), variable (XP)	Serum, heparinized plasma, EDTA plasma, citrate plasma	Antibodies (unspecified)	FDA CE-IVD	CLIA, fully automated
Siemens Enzygnost Syphilis Test (BEP 2000 Advance) 	Qualitative	4 plates per run; low-to-medium throughput	N/A	Serum, plasma	IgG or IgM	CE-IVD	EIA; microtitration plate analyser, photometric reading
Siemens Enzygnost Syphilis Test (BEP III) 	Qualitative	10 plates per run (continuous loading); high throughput	N/A	Serum, plasma	IgG or IgM	CE-IVD	EIA
Siemens IMMULITE Syphilis Screen Test and Siemens IMMULITE 2000 (automated through VersaCell) 	Qualitative	200 tests per hour; menu of more than 1000 assays	(5 hour walkaway time)	Serum, plasma, urine	Antibodies (unspecified)	FDA CE-IVD	CLIA
Siemens IMMULITE 2000 XPi (automated through VersaCell) 	Qualitative	200 tests per hour	35 minutes (5 hour walkaway time)	Serum, plasma, urine	Antibodies (unspecified)	FDA CE-IVD	CLIA

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CLEIA: chemiluminescence enzyme immunoassay; CLIA: chemiluminescence immunoassay; CMIA: chemiluminescent microparticle immunoassay; ECL: electrochemiluminescence; ECLIA: electrochemiluminescence immunoassay; EDTA: ethylenediaminetetraacetic acid; EIA: enzyme immunoassay; FDA: U.S. Food and Drug Administration; IgG: immunoglobulin G; IgM: immunoglobulin M; MFIA: multiplexed flow immunoassay; N/A: not applicable.

ARCHITECT Syphilis (Abbott): The ARCHITECT Syphilis TP assay from Abbott is a two-step immunoassay for the qualitative detection of antibody to *T. pallidum* in human serum or plasma using chemiluminescent microparticle immunoassay (CMIA) technology with flexible assay protocols, referred to as Chemiflex.

The assay can be run on the ARCHITECT $i1000_{SR}$, $i2000_{SR}$ or *i*4000 SR systems. In the first step of the assay, sample, microparticles coated with recombinant *T. pallidum* antigens (TpN15, TpN17 and TpN47) and assay diluent are combined. Anti-treponemal antibodies present in the sample bind to the *T. pallidum*-coated microparticles. After washing, the acridinium-labelled anti-human IgG and IgM conjugate is added in the second step. After

another wash cycle, pre-trigger and trigger solutions are added to the reaction mixture. The resulting chemiluminescent reaction is measured as relative light units (RLUs). A direct relationship exists between the amount of anti-treponemal antibodies in the sample and the RLUs detected by the ARCHITECT *i* optical system. The presence or absence of anti-treponemal antibodies in the specimen is determined by comparing the chemiluminescent signal in the reaction to the cut-off signal determined from a previous ARCHITECT Syphilis TP calibration. If the chemiluminescent signal in the specimen is greater than or equal to the cut-off signal, the specimen is considered reactive for anti-*T. pallidum*.

The ARCHITECT *i*1000$_{SR}$, *i*2000$_{SR}$ and *i*4000$_{SR}$ systems are a family of immunoassay analysers that use Chemiflex technology. All systems can accommodate serum, plasma, whole blood and urine as samples. The ARCHITECT *i*1000$_{SR}$ is an automated random-access system. It is the most compact of the immunoassay analysers offered by Abbott. It can accommodate 65 samples at a time and can perform up to 100 tests per hour. It is well suited for low-to-medium-volume laboratories. Same-day testing and treatment (STAT) results can be performed in less than 20 minutes.

The ARCHITECT *i*2000$_{SR}$ immunoassay analyser is fully automated and offers a maximum throughput of up to 200 tests per hour. First test results are available in approximately 28 minutes. The ARCHITECT *i*2000$_{SR}$ can accommodate 135 samples with 35 priority and 100 routine areas; it has 25 refrigerated reagent positions for up to 12 500 tests. The ARCHITECT *i*2000$_{SR}$ is best suited to a medium-volume laboratory. For high-volume laboratories, the ARCHITECT *i*4000$_{SR}$ system, which consists of two ARCHITECT *i*2000$_{SR}$ analysers, is available.

BioPlex Syphilis IgG (Bio-Rad Laboratories): The BioPlex Syphilis IgG kit from Bio-Rad Laboratories is a MFIA intended for the qualitative detection of *T. pallidum* IgG antibodies in human serum. The assay can be run on the BioPlex 2200 multiplexing platform described earlier in this report.

BioPlex 2200 immunoassays use heterogeneous (multiplex) sets of magnetic beads to simultaneously detect multiple analytes. The beads are infused with varying ratios of two fluorescent classification dyes, creating unique bead sets. Beads in each set are coated with a ligand (i.e. antigen, antibody, analyte, etc.) specific to a particular assay, allowing the capture and detection of specific analytes from a sample. Target analytes are captured on bead surfaces and probed with a corresponding fluorescent conjugate. With excitation and emission spectra distinct from those of the classification dyes used to identify analyte and control beads, the conjugate serves as the "reporter" fluorescence signal.

After the reaction, bead fluorescence is measured by the instrument in a manner similar to the way fluorescently labelled cells are evaluated in a standard flow cytometer. The beads flow single-file through a flow cell in a sheath of fluid to be interrogated by two lasers. As each bead travels through the interrogation zone, light scatter and fluorescence are measured. Signals from one laser are used to determine bead classification (specific analyte or control), and signals from the other laser are used to measure reporter fluorescence (sample reactivity). System software converts the reporter signal to a relative fluorescence intensity (RFI) value, which is then converted to a fluorescence ratio using the inherent reporter fluorescence of the internal standard bead. The RFI is compared to an assay-specific calibration curve to determine analyte concentration in antibody index or other appropriate units, or to score the result qualitatively as reactive, equivocal or non-reactive.

LIAISON Treponema Assay (DiaSorin): The LIAISON Treponema Assay from DiaSorin uses chemiluminescence immunoassay (CLIA) technology for the qualitative detection of total antibodies of any class (IgG or IgM) directed against *T. pallidum* in human serum. Recombinant antigens specific for *T. pallidum* are used to coat the magnetic particles and are used in the tracer when linked to an isoluminol derivative (isoluminol–antigen conjugate). During the first incubation step, antibodies present in the calibrators, samples or controls bind to the solid phase. During the second incubation, the antibody conjugate reacts with the antibodies already bound to the solid phase. After each incubation, the unbound material is removed with a wash cycle. Subsequently, the starter reagents are added and a flash chemiluminescence reaction is induced. The light signal, and hence the amount of isoluminol–antigen conjugate, is measured by a photomultiplier as RLUs and is indicative of total antibodies to *T. pallidum* present in calibrators, controls or samples.

The LIAISON Treponema Assay can be run on any of a family of analysers from DiaSorin, that is, LIAISON, LIAISON XL and LIAISON XS. All systems use a "flash" CLIA technology with paramagnetic microparticle solid phase. This means there is compatibility among the systems. Each analyser takes serum, plasma, urine or cerebrospinal fluid samples. The LIAISON has flexible operating modes (random access, batch and STAT) with a broad assay menu, which leads to high efficiency due to the system's 6-hour walkaway time (or 720 tests). There are 144 primary sample tubes of varying size in racks of 12, each with continuous reloading. Fifteen assays are available, allowing up to 1500 determinations, with continuous reloading. Throughput for the system is up to 180 tests per hour with a time to first results of 17 minutes, dependent on the assay. All assay steps and incubations are performed by the LIAISON, except for the initial magnetic particle resuspension.

The LIAISON XL Analyzer is a fully automated and compact system, performing complete sample processing (sample predilutions, sample and reagent dispensing, incubations, wash processes, etc.), and measurement and evaluation. The LIAISON XL offers random access, batch or STAT processing. Throughput is 180 tests per hour, with time to first results of 17 minutes, depending on the assay. Like the LIAISON Analyzer, walkaway time

is approximately 6 hours. The system offers continuous loading of samples, reagents and consumables.

Finally, the LIAISON XS Analyzer is a single-system, completely automated benchtop analyser with an integrated touchscreen personal computer, and more compact than the LIAISON or LIASON XL analyser. A total of 48 specimen tubes can be loaded, among primary or secondary tubes, or calibrators or controls. The LIAISON XS sample area allows up to four sample racks simultaneously. The throughput of the instrument is 85 tests per hour, with 17 minutes to the first results, depending on the assay. The walkaway time is approximately 2 hours.

Lumipulse G TP-N (Fujirebio): The Lumipulse G TP-N from Fujirebio is a chemiluminescence enzyme immunoassay (CLEIA) for the qualitative determination of antibodies (IgG and IgM) to *T. pallidum* in human serum and plasma. The assay is based on CLEIA technology; it uses a two-step sandwich immunoassay method and can be run on the LUMIPULSE G1200 immunoassay analyser. The critical reagents consist of ferrite microparticles coated with recombinant *T. pallidum* antigens (Tp15–Tp17 and TpN47), a conjugate of the antigens labelled with alkaline phosphatase (ALP) and a substrate solution. The anti-*T. pallidum* antibodies in the specimen bind with the antigens coated on the particles to form immunocomplexes. A washing step follows, which removes any unbound materials. A second reaction is initiated by adding the conjugated (ALP-labelled) antigens to form additional complexes. A second wash cycle removes any unbound material and a substrate solution is added initiating a chemical reaction with emission of a luminescent signal. The luminescence (at 477 nm) is measured by a photometric detector. The amount of antibody in a specimen is automatically calculated from the cut-off value, based on the calibration data. The results are reported as cut-off index values and interpreted as reactive or nonreactive. The LUMIPULSE G1200 is an automated immunoassay analyser capable of measuring the chemiluminescence generated by the chemical reaction of antibodies in patient specimens binding to antigens in the test reagents. The instrument processes 120 tests per hour and provides results in 30 minutes for all assays. It also allows for continuous loading and unloading of samples, reagents and consumables without operational interruption.

Elecsys Syphilis (Roche): The Elecsys Syphilis assay from Roche is an electrochemiluminescence immunoassay (ECLIA) for the in vitro qualitative determination of total antibodies against *T. pallidum* in human serum and plasma samples. The assay can be run on any of three Roche cobas immunoassay analysers, the e411, e602 or e801. The samples are incubated with a mixture of biotinylated and ruthenylated thermostable recombinant TpN15, TpN17 and TpN47 antigens. The presence of the corresponding antibodies subsequently leads to the formation of double-antigen sandwich immune complexes. The addition of streptavidin-coated paramagnetic microparticles causes the immune complexes to bind to the solid phase due to biotin–streptavidin interactions. The microparticles are then magnetically captured on the electrode surface and any unbound material is removed. Chemiluminescence is then induced by applying voltage and measured with a photomultiplier. The total assay time is 18 minutes and the results are calculated automatically by the analyser software.

Each of the cobas e411, e601, e602 and e801 instruments from Roche is a fully automated analyser that uses electrochemiluminescence (ECL) technology for immunoassay analysis. They are designed for both quantitative and qualitative in vitro assay determinations of a broad range of applications. All can accommodate serum, plasma and urine samples; the e801 can also accommodate cerebrospinal fluid, supernatant and whole blood.

The cobas e411 analyser is the most compact of the three analysers. Up to 18 assays can be run at a throughput of up to 86 tests per hour. In addition, 9-minute STAT applications for emergency sample assays can also be run. The system features continuous random-access and flexible STAT priority settings. The e411 analyser is a module in the larger cobas e4000 series, which also includes the c311 for clinical chemistry. The cobas e4000 series is low-throughput, with the capacity to run up to 530 tests per hour, depending on system configuration.

The cobas e601 can run up to 25 assays at a throughput of up to 170 tests and includes 9-minute STAT applications for emergency sample assays. Like the e411 module, the e601 system features continuous random-access and flexible STAT priority settings.

The e601 analyser is a module in the larger cobas e6000 series, which consists of a core unit and a combination of up to three analytical modules, including the cobas e601 and the cobas c501 for clinical chemical testing. The core unit allows for continuous loading and unloading of samples. The loading capacity is 150 samples in two trays of 75. The core module offers a single control unit for all associated modules. The cobas c501 clinical chemistry module is a fully automated, medium-throughput module that performs photometric assay tests for a wide range of analytes.

The cobas e602 analyser is a mid-volume throughput immunoassay module that can perform a broad range of heterogeneous immunoassay tests. It can run up to 25 assays at a throughput of up to 170 tests per hour. It also allows for STAT testing. The cobas e602 analyser is a module in the larger cobas e8000 series.

Finally, the cobas e801 analyser offers more than 1000 immunoassays across a wide range of disease areas. The instrument allows for continuous loading of reagents and consumables and has a high uptime, while requiring less hands-on time. The cobas e801 is designed for high-volume immunoassay testing; it can perform up to 300 tests per hour via 48 reagent channels. No reagent preparation is required. Furthermore, up to four cobas e801 models can be configured in a series delivering up to 1200 tests per hour across up to 192 reagent positions.

The cobas e602 and e801 analysers are modules in the cobas e8000 modular analyser series. In addition to those modules, the cobas e8000 consists of a core unit, an ion-selective electrode module, a sample buffer module and three possible clinical chemistry analytical units. Overall, the cobas e8000 system is designed for high-throughput laboratories and offers 440 configurations to maximize laboratory capacity. The maximum throughput of the system is 9800 test per hour per configuration.

ADVIA Centaur Syphilis (Siemens): The ADVIA Centaur Syphilis assay is an IVD immunoassay for the qualitative determination of antibodies to *T. pallidum* in human serum or plasma (ethylenediaminetetraacetic acid [EDTA], lithium- or sodium-heparinized, sodium citrate) using the ADVIA Centaur XP and CP systems as an aid in the diagnosis of syphilis. The ADVIA Centaur syphilis assay is a fully automated, antigen sandwich assay that uses direct chemiluminometric technology. The ancillary pack reagent-containing acridinium-ester-labelled *T. pallidum* recombinant antigens is added to the sample. These *T. pallidum* antigens complex with the antibodies in the sample. The solid phase containing biotinylated *T. pallidum* recombinant antigens preformed to streptavidin-coated magnetic latex particles is then added to the sample. These particles capture the *T. pallidum* antigen–antibody complexes. Antibody–antigen complexes will form if syphilis antibodies are present in the sample.

The ADVIA Centaur instruments are a family of automated immunoassay systems. According to the manufacturer, both systems are designed for ease of use. The ADVIA Centaur CP System is a medium-volume, high-throughput benchtop system. The system has a broad assay menu and fast turnaround time of 15 minutes for most assays. The system can run up to 180 tests per hour, with a 400-test walkaway capacity. It also provides uninterrupted system processing when loading samples, reagents and consumables.

The ADVIA Centaur XP System is a high-volume, high-performance system with more on-board reagents and dedicated STAT capabilities than the ADVIA Centaur CP System. The ADVIA Centaur XP System can run up to 240 tests per hour; it is always ready and has continuous operation without interruption. The productivity of the ADVIA Centaur XP System can also be enhanced by connecting it to other Siemens solutions, for example, Aptio Automation, ADVIA Automation, StreamLAB Automation or VersaCell solutions.

Enzygnost Syphilis (Siemens): The Enzygnost Syphilis test from Siemens, which is CE-IVD-marked, is an EIA for the qualitative detection of specific antibodies to *T. pallidum* in human serum or plasma. It is a competitive one-step EIA. *T. pallidum*-specific antibodies (IgG or IgM) contained in the sample and the peroxidase (POD)-labelled antibodies (anti-*T. pallidum*–POD conjugate) compete for binding to the *T. pallidum* antigens coated onto the wells of the microtitration plate.

The enzyme portion of the conjugate causes the chromogen working solution to turn blue. This reaction is stopped by adding the POD stopping solution, which causes a colour change to yellow. The intensity of the yellow colour produced is inversely proportional to the activity of *T. pallidum*-specific antibodies contained in the sample.

The Enzygnost Syphilis test is designed to be processed using the BEP III or BEP 2000 Advance systems from Siemens. The BEP 2000 Advance System is a fully automated, sample-to-result microtitration plate (MTP) analyser, which is designed for low-to-medium-volume laboratories. The system processes up to four MTPs in one run and allows reloading of one additional MTP. It records positive and negative events through process control and also provides photometric reading evaluation of the results. The BEP III System is also fully automated, including incubation, reagent dispensing, washing and photometric reading, as well as automated validation and evaluation. It is a high-throughput instrument running up to 10 MTPs in parallel with continuous MTP loading. Twenty different reagents can be loaded on the instrument. As such, the system is intended for use in high-volume laboratories.

IMMULITE 2000 Syphilis Screen (Siemens): The IMMULITE 2000 Syphilis Screen test is a treponemal testing procedure for the qualitative detection of antibodies to *T. pallidum* in human serum or heparinized plasma on the IMMULITE 2000 or IMMULITE 2000 XPi immunoassay system as an aid in the diagnosis of syphilis. The IMMULITE 2000 Syphilis Screen test is a solid-phase, one-step CLIA. The solid phase (beads) is coated with purified recombinant *T. pallidum* p17 (Tp17) antigen. The liquid phase consists of ALP (bovine calf intestine) conjugated to purified recombinant *T. pallidum* p17 (Tp17) antigen. The patient sample and reagent are incubated together with the coated beads for 30 minutes. During this time, total antibody to *T. pallidum* in the sample forms the antigen sandwich complex with purified recombinant *T. pallidum* p17 (Tp17) antigen on the bead and enzyme-conjugated purified recombinant *T. pallidum* p17 (Tp17) antigen in the reagent. The unbound patient sample and enzyme conjugate are then removed using centrifugal washes. Finally, chemiluminescent substrate is added to the reaction tube containing the beads and the signal is generated in proportion to the bound enzyme.

Both the IMMULITE 2000 and IMMULITE 2000 XPi immunoassay systems are automated and are designed for use in medium-to-high volume laboratories. Both offer large assay menus and can accommodate serum, plasma and urine samples, depending on the assay.

The IMMULITE 2000 system has a throughput of 200 tests per hour, with 5 hours of walkaway time. It has 24 resident assays, and a menu of more than 1000 assays can be performed. It can be automated through VersaCell or other ADVIA automation systems from Siemens. VersaCell, for example, is a compact, robotic system that connects two stand-alone instruments in flexible configurations. The IMMULITE 2000 immunoassay system also features intuitive software and a graphical user interface; it also offers streamlined information management, from remote test ordering to analysis of test results.

The IMMULITE 2000 XPi is a continuous random-access immunoassay analyser with a maximum throughput of 200 tests per hour and time to first results of 35 minutes. Walkaway time is up to 5 hours. It offers one of the largest immunoassay test menus available. The system has been enhanced with new workflow efficiency and productivity features, including an AutoStart feature that automates daily maintenance and an automated rack loader that enables users to load patient samples without pausing the system. The IMMULITE 2000 XPi system also delivers Internet-based remote monitoring capabilities and an enhanced touchscreen interface. Like the IMMULITE 2000 immunoassay system, the IMMULITE 2000 XPi can be automated through VersaCell.

In addition to the integrated assays and platforms manufactured by a single manufacturer, some *T. pallidum* assays have been developed for use on automated instruments manufactured by other companies. These are shown in **Table 3**.

Table 3: Treponemal systems using automated instruments

Manufacturer/test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Fujirebio SERODIA TPPA Auto (Beckman Coulter PK7000 series) OR Fujirebio SERODIA TPPA (manual)	N/A	N/A	N/A	Serum, plasma	Antibodies (unspecified)	CE-IVD	Particle agglutination, automated interpretation
Newmarket Biomedical NewBio TPHA and pk TPHA Assays (manual or uses the Beckman Coulter platforms: PK7300 and PK7400 Automated Microplate Systems) 	Qualitative and semi-quantitative	300 per hour; high throughput	20–60 minutes	Serum, plasma, red blood cells	IgG and IgM antibodies	FDA	Passive particle agglutination, charge-coupled device camera
Trinity Biotech CAPTIA Syphilis-G Assay (variety of compatible devices; manual, automatic, semi-automatic)	Qualitative	N/A	N/A	Serum	IgG antibodies	FDA CE-IVD	EIA, spectrophotometer
ZEUS Scientific AtheNA Multi-Lyte *T. pallidum* IgG Plus Test System (platform: Luminex 100/200 System) 	Qualitative	Multiplex, up to 100 analytes	≤45 minutes	Serum	IgG antibodies	FDA CE-IVD	MFIA, open platform, flow cytometry

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; EIA: enzyme immunoassay; FDA: U.S. Food and Drug Administration; IgG: immunoglobulin G; IgM: immunoglobulin M; MFIA: multiplexed flow immunoassay; N/A: not applicable.

SERODIA TPPA and SERODIA TPPA Auto (Fujirebio): The SERODIA TPPA and SERODIA TPPA Auto test are based on Fujirebio's particle agglutination assay using gelatin particles coated with purified pathogenic *T. pallidum* (Nichols strain). These coated gelatin particles are agglutinated in the presence of antibodies to *T. pallidum* in human serum and plasma. The SERODIA TPPA test can be performed manually, while the SERODIA TPPA Auto can be performed using the PK7000 series automated microplate systems. Serum or plasma samples are serially diluted in sample diluent in microplate wells. Sensitized gelatin particles are added to the respective wells and the contents of the plate are mixed by hand or on a tray mixer. Serum or plasma containing specific antibodies will react with the antigen-sensitized coloured gelatin particles to form a smooth mat of agglutinated particles in the microtitration tray. A compact button formed by the settling of the non-agglutinated particles characterizes negative reactions. The agglutination patterns and interpretation of the test are clear-cut and easy to read visually or with the aid of a tray viewer.

NewBio TPHA and pk TPHA Syphilis Assays (Newmarket Biomedical): The NewBio TPHA and pk TPHA Syphilis assays are passive particle agglutination assays for the qualitative and semi-quantitative detection of IgG and IgM antibodies to *T. pallidum*. They are based on the principle of agglutination and pattern recognition. The tests use preserved avian erythrocytes sensitized with extracted antigens of *T. pallidum* (Nichols strain). Specific antibodies present in an EDTA plasma or serum test sample bind to these antigens when the sample is incubated with the particles. This causes the particles to agglutinate, then settle to form a characteristic pattern in the test well. Visually, a reactive test is a homogenous layer of cells. A nonreactive test results in a compact dense button surrounded by a clear zone.

Both the NewBio TPHA and pk TPHA tests can be run and interpreted manually or with an auto-analyser using an agglutination interpretation programme. In addition, the NewBio pk TPHA test can be performed on the PK7300 and PK7400 Automated Microplate Systems from Beckman Coulter. Both systems are high throughput (300 samples per hour, with a 20–60-minute reaction time), fully automated batch systems. They can accommodate plasma, serum, red blood cells and use agglutination technology with Beckman Coulter terraced microplates.

The PK7400 is the fourth generation of the PK7000 series of analysers, all of which use the same testing methodologies. System controls include process controls, such as reagent volume monitoring, clot detection and a 30 °C incubation area that is monitored throughout testing. The PK7400 reads the settling patterns of the erythrocytes in each well based on the threshold settings chosen for each reagent. It captures the well images using a charge-coupled device camera and subsequently uses the threshold settings in its algorithm to differentiate agglutinated from non-agglutinated patterns.

CAPTIA Syphilis-G (Trinity Biotech): The CAPTIA Syphilis-G assay is an enzyme immunoassay for the qualitative detection of IgG antibodies to *T. pallidum* in serum specimens, to be used in conjunction with non-treponemal testing to provide serological evidence of infection with *T. pallidum* (the agent of syphilis). CAPTIA Syphilis-G may be run manually or may be used with a variety of automatic or semi-automatic processors and liquid handling systems.

The test procedure is as follows. Microtitration wells coated with *T. pallidum* antigens are exposed to test specimens that may contain specific antibodies. After an incubation period, unbound components in the test sample are washed away. Specifically bound IgG reacts with a conjugated horseradish peroxidase monoclonal antibody during a second incubation period. After a second wash cycle, specifically bound enzyme conjugate is detected by reaction with tetramethylbenzidine. The enzymatic reaction is stopped using 1N sulphuric acid. The assay is measured spectrophotometrically to indicate the presence or absence of IgG treponemal antibodies.

AtheNA Multi-Lyte T. pallidum IgG Plus Test System (ZEUS Scientific): The AtheNA Multi-Lyte *T. pallidum* IgG Plus Test System, is an MFIA intended for the qualitative detection of specific human IgG class antibodies to *T. pallidum* in human serum. It uses traditional sandwich immunoassay techniques. The platform for this assay is the Luminex 100/200 System.

The AtheNA Multi-Lyte *T. pallidum* IgG Plus test procedure involves two incubation steps where patient sera are diluted and the diluted test sera are incubated in a vessel containing a multiplexed mixture of the bead suspension. The multiplexed bead suspension contains a mixture of distinguishable sets of polystyrene microspheres; each set is conjugated with *T. pallidum* antigen. The bead mix also contains one bead set designed to detect nonspecific antibodies in the patient sample (if present); four separate bead sets are used for assay calibration. If present in patient sera, the individual antibodies will bind to the corresponding immobilized antigen bead set.

The Luminex 100/200 System is a flexible, automated analyser based on the principles of flow cytometry. The system will multiplex up to 100 analytes in a single 96-well plate, using very small samples. Read time is less than 45 minutes. This is an open platform consisting of solid-phase microparticles on which the immunoassays are built; a modified flow cytometer used to interpret the reactions on the microparticles.

The Luminex 100/200 System consists of three xMAP Technologies. The first is xMAP microspheres, a family of fluorescently dyed micrometre-sized polystyrene microspheres that act as both the identifier and the solid surface to build the assay. The second is a flow cytometry-based instrument, the Luminex 100/200 analyser, which integrates key xMAP detection components, such as lasers, optics, fluidics and high-speed digital signal processors. Finally, the third component is the xPONENT software, which is designed for protocol-based data acquisition with robust data regression analysis.

Finally, there are several *T. pallidum* assays that are intended for manual use. These are shown and described in **Table 4**.

ENZY-WELL SYPHILIS IgG (Diesse Diagnostics): The ENZY-WELL SYPHILIS IgG test from Diesse Diagnostics is an immunoenzymatic method for the qualitative detection of IgG antibodies to *T. pallidum* in human serum and plasma. The test is based on the enzyme-linked immunosorbent assay (ELISA) technique. The diluted patient sample is incubated in microplate wells coated with *T. pallidum*. During this incubation, specific immunoglobulins if present, bind to the antigen in the well. After washing to eliminate unbound proteins, a second incubation is performed with the conjugate, composed of human IgG monoclonal antibodies labelled with POD. After washing to remove unbound conjugate from the wells, the substrate is added, which reacts to produce colour in the presence of the POD. An acidic solution is added to stop the reaction and the absorbance of the developed colour is read at 450 nm.

TREP-SURE EIA (Phoenix Biotech, a subsidiary of Trinity Biotech): The TREP-SURE EIA is a qualitative enzyme immunoassay for the IVD detection of *T. pallidum* antibodies in human serum or EDTA and citrated plasma. This product can be used as an initial screening test or as a confirmatory diagnostic test. Microtitration wells coated with *T. pallidum* antigens are exposed to test specimens that may contain specific antibodies. After an incubation period, unbound components in the test sample are washed away. Specifically bound IgG reacts with a conjugated horseradish peroxidase monoclonal antibody during a second incubation period. After a second wash cycle, specifically bound enzyme conjugate is detected by reaction with tetramethylbenzidine.

The enzymatic reaction is stopped using 1N sulphuric acid. The assay is measured spectrophotometrically to indicate the presence or absence of IgG treponemal antibodies.

Immunofluorescence Assay FTA Absorption (FTA-ABS) Test System (ZEUS Scientific): The ZEUS FTA-ABS Test System is designed for the qualitative determination of antibodies to *T. pallidum* and is intended to be used as an aid in the confirmation of syphilis antibodies. The FTA-ABS Test System uses nonviable *T. pallidum* (Nichols strain) cells as a substrate (antigen). The reaction occurs in two steps: *(1)* the substrate cells are reacted with specially treated patient sera. If treponemal antibodies are present in the patient sera, an antigen–antibody reaction takes place between the substrate cells and the circulating anti-treponemal antibodies in the patient sera; *(2)* goat anti-human immunoglobulin labelled with fluorescein isothiocyanate is added to the *T. pallidum* substrate cells. The substrate cells are then examined with a fluorescence microscope. The intensity of staining is graded on a scale of 1+ to 4+ or as negative (no fluorescence).

Table 4: Treponemal assays (manual)

Manufacturer/ test	Reporting	Sample type	Detection	Approval status	Test technology
Diesse Diagnostics ENZY-WELL SYPHILIS IgG	Qualitative	Serum, plasma	IgG	FDA CE-IVD	ELISA
Phoenix Biotech TREP-SURE EIA	Qualitative	Serum, plasma	IgG	FDA CE-IVD	EIA, spectrophotometer
ZEUS Scientific FTA-ABS Test System	Qualitative	Serum	IgG, IgM	FDA CE-IVD	Fluorescence microscope

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; EIA: enzyme immunoassay; ELISA: enzyme-linked immunosorbent assay; FDA: U.S. Food and Drug Administration.

3.2 Rapid diagnostic testing for syphilis

In recent years, a range of RDTs for syphilis screening has been developed. Virtually all of these tests are antibody tests that detect *T. pallidum*. However, there is now at least one near-POC non- treponemal test. However, because of the persistence of treponemal antibodies, these *T. pallidum* RDTs cannot distinguish between active and past treated infections. However, in resource-limited settings, where many people do not have access to laboratory-based non-treponemal tests to confirm active syphilis, pregnant women who seropositive with a *T. pallidum* RDT are treated for syphilis to prevent transmission of the infection. As indicated by Jafari et al.: "This is now accepted practice as the risk of over-treatment due to biological false positives which are not syphilis in origin is more acceptable than the risk of non-treatment of syphilis" *(41)*.

Commercially available RDTs that detect *T. pallidum* are summarized in **Table 5** and listed alphabetically according to manufacturer.

Table 5: Commercially available RDTs for *Treponema pallidum* detection

Manufacturer/ test	Sample type	Sample volume	Turnaround time	Detection	Approval status	Test type
Abbott Determine Syphilis TP	Whole blood (fingerstick), plasma, serum	50 µl	15 minutes (up to 24 hours)	N/A	CE-IVD	Lateral flow strip
Abbott SD Bioline Syphilis 3.0	Whole blood (venous or fingerstick), plasma, serum	20 µl (whole blood), 10 µl (plasma or serum)	5–20 minutes	IgG, IgM, IgA	CE-IVD	Cassette-enclosed test card
AccuBio Tech Accu-Tell Syphilis Rapid Test	Plasma, serum	150 µl (strip), 40 µl (cassette)	5–15 minutes	IgG, IgM	CE-IVD	Lateral flow strip, cassette
CTK Biotech OnSite Syphilis Ab Combo Rapid Test	Whole blood (venous or fingerstick)	One drop	15 minutes	IgG, IgM and IgA	CE-IVD	Cassette-enclosed test card
Cypress Diagnostics Syphilis Rapid Test	Whole blood, plasma, serum	20 µl (whole blood), 10 µl (plasma or serum)	5–20 minutes	N/A	N/A	Cassette-enclosed test card
MedMira Reveal TP	Whole blood (venous or fingerstick), plasma, serum	One drop (fingerstick), five drops (venepuncture), one drop (serum or plasma)	Immediately	N/A	CE-IVD	Cassette
Omega Diagnostics VISITECT SYPHILIS	Whole blood (venous or fingerstick), plasma, serum	50 µl	30 minutes	IgG, IgM	CE-IVD	Cassette-enclosed test card
Premier Medical Corporation First Response Syphilis Anti-TP Card	Whole blood (venous or fingerstick), plasma, serum	75 µl	15–25 minutes	IgG, IgM	WHO prequalification	Cassette-enclosed test card
Trinity Biotech Syphilis Health Check	Whole blood (venous or fingerstick), plasma, serum	Two drops	10 minutes	N/A	FDA	Cassette-enclosed test card
Trinity Biotech Uni-Gold Syphilis Treponemal	Whole blood (venous or fingerstick), plasma, serum	Approximately 60 µl	Approximately 15 minutes	N/A	CE-IVD	Cassette-enclosed test card

Table 5 (continued): Commercially available RDTs for *Treponema pallidum* detection

Manufacturer/ test	Sample type	Sample volume	Turnaround time	Detection	Approval status	Test type
The Tulip Group/Qualpro Syphicheck-WB	Whole blood (venous or fingerstick), plasma, serum	25 µl	15 minutes	IgG, IgM	CE-IVD	Cassette-enclosed test card

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: U.S. Food and Drug Administration; IgG: immunoglobulin G; IgM: immunoglobulin M; MFIA: multiplexed flow immunoassay; N/A: not applicable; WHO: World Health Organization.

Near-POC non-treponemal test

The Abbott IMPACT RPR is a near-POC non-treponemal test that is read by the naked eye and requires instrumentation (microscope). However, it requires serum or plasma as a sample. The IMPACT RPR test kit is for the detection of non-treponemal RPR antibodies to assist in the diagnosis of syphilis. The test turnaround time is 8 minutes. Abbott indicates that the performance of the test is consistent with an FTA absorption test.

Treponemal and non-treponemal test

A combination treponemal/non-treponemal test can be used to diagnose syphilis at the POC where traditional laboratory-based testing may not be available. The only commercially available dual non-treponemal and treponemal syphilis test for use at POC is summarized in **Table 6**.

DPP Syphilis Screen and Confirm Assay (Chembio Diagnostic Systems): Chembio developed the first dual non-treponemal and treponemal syphilis test for use at the POC. The assay uses a unique combination of protein A and anti-human IgM antibody, which are conjugated to colloidal gold particles. It also uses a recombinant antigen to *T. pallidum* and synthetic antigens for non-treponemal testing, separately bound to the membrane solid phase. The result is an assay that permits the simultaneous, yet separate detection of both markers. The DPP Syphilis Screen and Confirm Assay requires a sample size of only 10 µl of whole blood (fingerstick or venepuncture), and tests can be stored at room temperature (2–30 °C). The turnaround time of the test is 15–20 minutes. The test is not only highly sensitive and specific, but also useful for the serological diagnosis of syphilis in primary health care clinics or resource-poor settings.

Combined HIV/syphilis tests currently on the market

This section of the report describes the available combined HIV and syphilis tests designed for use at the POC, all of which are RDTs. The available tests are summarized in **Table 7** and alphabetized according to the manufacturer.

Table 6: Combination treponemal and non-treponemal test

Manufacturer/test	Sample type	Sample volume	Turnaround time	Approval status
Chembio DPP Syphilis Screen and Confirm Assay 	**Whole blood (fingerstick or venepuncture)**	**10 µl**	**15–20 min**	**CE-IVD**

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device

Table 7: Combined HIV and syphilis RDT tests

Manufacturer/test	Sample type	Turnaround time	Test type
Abbott SD BIOLINE HIV/Syphilis Duo Test 	Whole blood (fingerstick or venepuncture)	15–20 minutes	Lateral flow
bioLytical Laboratories INSTI HIV/Syphilis Multiplex Test 	Serum	60 seconds	Immunofiltration
Chembio DPP HIV-Syphilis System 	Whole blood (fingerstick or venepuncture), serum, plasma	10 minutes	Immunochromatography
CTK Biotech OnSite HIV/Syphilis Ab Combo Rapid Test 	Whole blood, serum, plasma	15 minutes	Lateral flow chromatography
MedMira Multiplo Rapid TP/HIV Antibody Test 	Serum	3 minutes	Immunofiltration
Premier Medical Corporation First Response HIV 1+2/Syphilis Combo Card Test	Whole blood, serum, plasma	15 minutes	Immunochromatography
SD Biosensor STANDARD Q HIV/ Syphilis Combo Test 	Serum, plasma, whole blood	15 minutes	Immunochromatography

Each of the seven combined HIV and syphilis assays currently on the market is described here.

SD BIOLINE HIV/Syphilis Duo Rapid Test (Abbott): The SD BIOLINE HIV/Syphilis Duo Rapid Test is an easy-to-use, rapid lateral flow assay for the simultaneous detection of HIV-1, including subtype O, and HIV-2 or syphilis *T. pallidum* from whole blood (venous or fingerstick), serum or plasma samples with results in approximately 15–20 minutes.

INSTI HIV/Syphilis Multiplex Test (bioLytical Laboratories): The INSTI HIV/Syphilis Multiplex Test is designed to provide rapid qualitative detection of HIV-1 and HIV-2 and syphilis *T. pallidum* in a rapid test format using immunofiltration. Turnaround time is about 60 seconds.

DPP HIV-Syphilis System (Chembio Diagnostic Systems): The DPP HIV-Syphilis System from Chembio is a single-use immunochromatographic, rapid screening test for the detection of antibodies both to HIV types 1 and 2 and syphilis *T. pallidum* in fingerstick whole blood, venous whole blood, serum or plasma samples. The test, which requires only 10 µl of blood, includes the Chembio SampleTainer specimen collection bottle, which is a safe closed system for handling potentially infectious blood samples. Turnaround time for the test is about 10 minutes.

OnSite HIV/Syphilis Ab Combo Rapid Test (CTK Biotech): The OnSite HIV/Syphilis Ab Combo Rapid Test assay is a lateral flow chromatographic immunoassay for the qualitative detection of antibodies to HIV-1, HIV-2 and *T. pallidum*. It is a three-line test that can be used with whole blood, serum or plasma to detect IgG, IgM and immunoglobulin A (IgA) to HIV-1 and HIV-2 and *T. pallidum*. Results are available within 15 minutes.

Multiplo Rapid TP/HIV Antibody Test (MedMira): The Multiplo Rapid TP/HIV Antibody Test from MedMira, is CE-IVD-marked, making it available for sale and distribution throughout the European Union; the company is pursuing product registration in a number of other markets. The combination assay is built on the MedMira rapid vertical flow technology platform and is sold in the same packaging formats as the company's rapid HIV antibody test. The Multiplo Rapid TP/HIV Antibody Test combines qualitative detection of HIV-1 and HIV-2 with qualitative detection of in an immunofiltration format. Turnaround time is approximately 3 minutes.

First Response HIV 1+2/Syphilis Combo Card Test (Premier Medical Corporation): The First Response HIV 1+2/Syphilis Combo Card Test is based on the principle of immunochromatography for the qualitative detection of antibodies (IgG and IgM) specific for HIV 1+2 or syphilis. It is a three-line test that can be used with whole blood, serum or plasma with results within 15 minutes.

STANDARD Q HIV/Syphilis Combo (SD Biosensor): The STANDARD Q HIV/Syphilis Combo test is a qualitative, lateral flow assay to detect antibodies specific to HIV-1, HIV-2 and syphilis (*T. pallidum*) in serum, plasma and whole blood using immunochromatography. Turnaround time is approximately 15 minutes.

3.3 Tests for detecting *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Trichomonas vaginalis*, *Mycoplasma genitalium*, herpes simplex virus 1 and 2 and HPV

The following sections present testing methods that are commonly used to identify the remaining STIs that are the subjects of this report, that is, immunoassays and molecular tests to detect *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV-1 and HSV-2, and HPV. These have been grouped together, separate from syphilis, because most are available in some combination on multiplex platforms. There are also more information and tests available for syphilis due to the critical need for interventions to eliminate mother-to-child transmission of HIV and syphilis.

Generally speaking, the primary methods used today to detect *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV-1 and HSV-2, and HPV are molecular tests; where available, commercial immunoassays are included. The commercial molecular platforms described here are automated and have single or multiplex assays for detecting one analyte per assay or multiple bacteria (and viral pathogens) *(37)*. Many of them have separate automated instruments to perform sample preparation and nucleic acid extraction. As mentioned, these platforms are also used across the multiple STIs considered here and will also be discussed as such in a separate section after the individual descriptions of each of the STIs and testing methods particular to each infection. This section introduces both molecular tests and platforms.

Lateral flow tests for *Chlamydia trachomatis*

The lateral flow antibody tests available for *C. trachomatis* are easy to use and relatively inexpensive lateral flow or RDT tests. These include, but are not limited to: ACON Chlamydia (ACON Laboratories); aQcare Chlamydia TRF kit (Medisensor); Biorapid Chlamydia Ag Test (Biokit); Chlamydia Rapid Test SAS (Diagnostics for the Real World); CLEARVIEW Chlamydia (Alere); Chlamydia Test Card (UltiMed); HandiLab-C (HandiLab); and QuickVue (QuidelOrtho). In a systematic review of the listed assays, Kelly et al. found that although *C. trachomatis* antigen detection rapid POC tests exhibited high specificity across all specimen types (range: 97–99%), pooled sensitivity was much lower (37–63%) *(42)*.

Molecular tests for *Chlamydia trachomatis*

Molecular testing is considered the gold standard for the detection of *C. trachomatis* and there are several molecular laboratory platforms available for this purpose. Most are combination assays also able to detect *N. gonorrhoeae*. Laboratory-based molecular platforms with assays that detect *C. trachomatis* are described in **Table 8**. There are also molecular-based platforms for use at POC or near-POC to detect *C. trachomatis* and these are included in **Table 9**. Molecular tests for *C. trachomatis* require either swab or urine samples (no blood-based samples).

Many of these assays detect other STIs, as shown in **Table 23**, but for the purposes of this section, we have focused on characteristics specific for *C. trachomatis*.

Table 8: Molecular laboratory-based platforms for the detection of *Chlamydia trachomatis*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Abbott Alinity m System (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, high-risk human papillomavirus) 	Qualitative	N/A	Less than 115 minutes	Endocervical swab, vaginal swab, urine, oropharyngeal, rectal	RNA	CE-IVD	RT–PCR
Abbott m2000 RealTime System (*C. trachomatis*, *N. gonorrhoeae* assay) 	Qualitative	96 samples per run	N/A	Symptomatic: endocervical and vaginal swab, male urethral swab, urine swab; asymptomatic: vaginal swab, urine swab	Plasma DNA	FDA	Real-time PCR
BD ProbeTec ET system (*C. trachomatis*/*N. gonorrhoeae*) 	Qualitative	46 samples	3 hours	Endocervical swab, male urethral swab, urine	DNA	FDA	SDA
BD Viper XTR system (ProbeTec CT Q^x, GC Q^x, TV Q^x, HSV-1 and 2 Q^x Amplified DNA assays) 	Qualitative	96 with continuous loading	40 minutes	Endocervical swab, male urethral swab; male and female urine specimens	DNA	FDA	SDA and rPCR

Table 8 (continued): Molecular laboratory-based platforms for the detection of *Chlamydia trachomatis*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
BD Viper LT System (ProbeTec CT Q^x, GC Q^x assays, and Onclarity HPV) 	Qualitative	30	3.5 hours	Endocervical swab, male urethral swab, vaginal swab, female urine	DNA	FDA	SDA and rPCR
BD MAX System (CT/GC/TV assay) 	Qualitative	24	2.5–3 hours	Male urine, female urine, endocervical swab, vaginal swab	DNA	FDA	Real-time PCR
BIONEER *ExiStation* Universal Molecular Diagnostic System (AccuPower assays *C. trachomatis*, *C. trachomatis* and *N. gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*/*M. genitalium*, *T. vaginalis*, *T. vaginalis*/HSV, HPV), with *ExiPrep* and *Exicycler* 	N/A	N/A	N/A	Urine, vaginal swab, urethral swab	DNA	CE-IVD (except C. trachomatis/ *N. gonorrhoeae*)	Real-time PCR
ELITechGroup Solutions STI ELITe MGB panel (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*) 	N/A	N/A	2.5 hours	Urine, cervical swab	DNA	CE-IVD	Real-time PCR
Hain Lifescience FluoroType system (*C. trachomatis*, *N. gonorrhoeae* assays), with GenoXtract and FluoroCycler PCR 	Quantitative	12	Varies by assay	Cervical swab, urine, ejaculate	DNA	CE-IVD	qPCR

Table 8 (continued): Molecular laboratory-based platforms for the detection of *Chlamydia trachomatis*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Hologic Panther System (Aptima assays – Combo 2 *C. trachomatis*/*N. gonorrhoeae*, herpes simplex viruses 1 and 2, HPV types 16 18/45 genotype, HPV, *Mycoplasma genitalium*, *Trichomonas vaginalis*) 	Qualitative	120	3 hours (first result), 5 minutes each thereafter	Endocervical swab, vaginal swab, gynaecological specimen, male urethral swab, rectal swab, urine, throat swab	rRNA	CE-IVD FDA	TMA, HPA
QIAGEN N.V. QIAsymphony artus kits (*C. trachomatis*/*N. gonorrhoeae*, QS- Rotor-Gene Q, HSV-1/2 Rotor-Gene Q), with SP/AS and Rotor-Gene Thermocyclers 	Qualitative	96	N/A	Vaginal swab, urine	Plasmid, genomic DNA	CE-IVD	Real-time PCR
QIAGEN N.V. digene tests (HC2 CT-GC Dual ID, HC2 CT-ID DNA, HC2 CT/GC, HC2 GC-ID, HPV) with Rapid Capture System 	Qualitative	352	In 8-hour shift	Cervical specimen	DNA	FDA	Chemilumi-nescence
QIAGEN N.V. NeuMoDx assays (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*), with NeuMoDx 288 and NeuMoDx 96  	Qualitative	N/A	N/A	Urine	DNA	CE-IVD	Real-time PCR

Table 8 (continued): Molecular laboratory-based platforms for the detection of *Chlamydia trachomatis*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Roche cobas 6800/8800 systems (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*) 	Qualitative	96	3.5 hours	Urine, vaginal swab, endocervical swab	DNA	CE-IVD FDA	PCR
Seegene Allplex, Anyplex and Seeplex test kits; open system (*C. trachomatis*/*N. gonorrhoeae*, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV, HPV) 	Quantitative	N/A	N/A	N/A	DNA	CE-IVD	Real-time PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: U.S. Food and Drug Administration; HPA: hybridization protection assay; N/A: not applicable; PCR: polymerase chain reaction; qPCR: quantitative PCR; rPCR: random PCR; rRNA: ribosomal RNA; RT–PCR: PCR with reverse transcription; SDA: strand displacement amplification; TMA: transcription-mediated amplification.

Note: The information in this table is specific for *Chlamydia trachomatis* although most assays detect other STIs; see **Table 22** in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Table 9: Molecular platforms for use at POC or near-POC for *Chlamydia trachomatis*

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
AmplexDiagnostics eazyplex Complete STDs (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*), eazyplex STD *C. trachomatis*, eazyplex STD *C. trachomatis*/*N. gonorrhoeae* (Genie III platform) 	Qualitative	N/A	Urethral, rectal/anal, vaginal, cervical and pharyngeal swab	CE-IVD	N/A
binx io *C. trachomatis*/*N. gonorrhoeae* assay (*io* system)  	Qualitative	30 minutes	Vaginal swab, male urine	FDA CE-IVD CLIA-waived	N/A
Bosch Healthcare Solutions STI Multiplex Array/ Vivalytic Analyzer (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV) 	Qualitative	2 hours 30 minutes	Urogenital swab, urine specimen	CE-IVD	Biochip array
Cepheid Xpert *C. trachomatis*/*N. gonorrhoeae* assay (GeneXpert System) 	Qualitative	90 minutes	Urine, endocervical swab, rectal swab, throat swab, vaginal swab	FDA CE-IVD	N/A
HiberGene Diagnostics HG *C. trachomatis*/*N. gonorrhoeae* Combo test (HG Swift and HG Swift Plus) 	Qualitative	<60 minutes for positive, 70 minutes for negative	Vaginal swab, urine	CE-IVD	LAMP

Table 9 (continued): Molecular platforms for use at POC or near-POC for *Chlamydia trachomatis*

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
Molbio Diagnostics Truenat *C. trachomatis*, *C. trachomatis*/*N. gonorrhoeae* assays (Truelab Real Time micro PCR System) 	Semi-quantitative	35 minutes	Endocervical swab, vaginal swab, male urethral swab, urine	CE-IVD	microPCR
Ustar Biotechnologies *C. trachomatis*/*N. gonorrhoeae* (CPA Assay) (EasyNAT Platform) 	Qualitative	50 minutes	Male urinary tract swab, cervical swab	N/A	CPA
Visby Medical *C. trachomatis*/*N. gonorrhoeae*/*T. vaginalis* test 	Qualitative	50 minutes	Vaginal swab	FDA CLIA-waived	RT–PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CLIA: Clinical Laboratory Improvement Amendments waiver; CPA: cross priming amplification; FDA: U.S. Food and Drug Administration; LAMP: loop-mediated isothermal amplification; N/A: not applicable; PCR: polymerase chain reaction; RT–PCR: PCR with reverse transcription.

Note: The information in this table is specific for *C. trachomatis* although most assays detect other STIs; see **Table 23** in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Lateral flow tests for *Neisseria gonorrhoeae*

Like *C. trachomatis*, current lateral flow and RDT diagnostic tests for *N. gonorrhoeae* often have specificities greater than 90%, while sensitivities are often 50% or lower.

Molecular tests for *Neisseria gonorrhoeae*

Molecular testing is considered the gold standard for the detection of *N. gonorrhoeae* and there are several molecular laboratory platforms available for this purpose. Most of these assays can also detect *C. trachomatis*. Laboratory-based molecular platforms with assays that detect *N. gonorrhoeae* are described in **Table 10**. There are also molecular-based platforms for use at POC or near-POC to detect *N. gonorrhoeae* and are included in **Table 11**.

Table 10: Molecular laboratory-based platforms for the detection of *Neisseria gonorrhoeae*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Abbott Alinity m System (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, high-risk HPV) 	Qualitative	N/A	Less than 115 minutes	Endocervical swab, vaginal swab, urine, oropharyngeal, rectal	DNA	CE-IVD	RT–PCR
Abbott RealTime m2000 system (*C. trachomatis*/*N. gonorrhoeae* assay) 	Qualitative	96 samples per run	N/A	Symptomatic: endocervical and vaginal swab, male urethral swab, urine swab; asymptomatic: vaginal swab, urine swab	Genomic DNA	FDA	Real-time PCR
BD ProbeTec ET system (*C. trachomatis*/ gonorrhoea) 	Qualitative	46 samples	3 hours	Endocervical swab, male urethral swab, urine	DNA	FDA	SDA
BD Viper XTR system (ProbeTec *C. trachomatis* Q^x, gonorrhoea Q^x, *T. vaginalis* Q^x, HSV-1 and 2 Q^x Amplified DNA assays) 	Qualitative	96 with continuous loading	40 minutes	Endocervical swab, male urethral swab; male and female urine specimens	DNA	FDA	SDA and rPCR
BD Viper LT system (ProbeTec *C. trachomatis* Q^x, gonorrhoea Q^x assays, and Onclarity HPV) 	Qualitative	30	3.5 hours	Endocervical swab, male urethral swab, vaginal swab, female urine; cervical specimen (HPV)	DNA	FDA	SDA and rPCR
BD MAX system (*C. trachomatis*/ gonococcus/*T. vaginalis* assay) 	Qualitative	24	2.5–3 hours	Male urine, female urine, endocervical swab, vaginal swab	DNA	FDA	Real-time PCR

Table 10 (continued): Molecular laboratory-based platforms for the detection of *Neisseria gonorrhoeae*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Bioneer ExiStation Universal Molecular Diagnostic System (AccuPower assays – *C. trachomatis*, *C. trachomatis* and *N. gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*/*M. genitalium*, *T. vaginalis*, *T. vaginalis*/HSV, HPV), with ExiPrep and Exicycler 	N/A	N/A	N/A	Urine, vaginal swab	DNA	CE-IVD (except *C. trachomatis*/*N. gonorrhoeae*)	Real-time PCR
ELITechGroup Solutions STI ELITe MGB panel (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*) 	N/A	N/A	2.5 hours	Urine, cervical swab	DNA	CE-IVD	Real-time PCR
Hain Lifescience FluoroType system (*C. trachomatis*, *N. gonorrhoeae* assays), with GenoXtract and FluoroCycler PCR 	Quantitative	12	Varies by assay	Cervical swab, urine	DNA	CE-IVD	qPCR
Hologic Panther System (Aptima assays – Combo 2 *C. trachomatis*/*N. gonorrhoeae*, herpes simplex viruses 1 and 2, human papillomavirus 16 18/45 genotype, HPV, *M. genitalium*, *T. vaginalis*) 	Qualitative	120	3 hours (first result), 5 minutes each thereafter	Endocervical swab, vaginal swab, gynaecological specimen, male urethral swab, rectal swab, urine, throat swab	rRNA	CE-IVD FDA	TMA, HPA

Table 10 (continued): Molecular laboratory-based platforms for the detection of *Neisseria gonorrhoeae*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
QIAGEN N.V. QIAsymphony artus kits (*C. trachomatis/N. gonorrhoeae* QS-Rotor-Gene Q, HSV-1/2 Rotor-Gene Q), with SP/AS and Rotor-Gene Thermocyclers 	Qualitative	96	N/A	Vaginal swab, urine (*C. trachomatis/N. gonorrhoeae*)	Genomic DNA	CE-IVD	Real-time PCR
QIAGEN N.V. digene tests (HC2 *C. trachomatis*-gonorrhoea Dual ID, HC2 *C. trachomatis*-ID DNA, HC2 *C. trachomatis*/ gonorrhoea, HC2 gonorrhoea-ID, HPV) with Rapid Capture System 	Qualitative	352	In 8-hour shifts	Cervical specimen	DNA	FDA	Chemilumi-nescence
QIAGEN N.V. NeuMoDx assays (*C. trachomatis/N. gonorrhoeae*, HPV, *T. vaginalis/M. genitalium*), with NeuMoDx 288 and NeuMoDx 96  	Qualitative	N/A	N/A	Urine	DNA	CE-IVD	Real-time PCR
Roche cobas 6800/8800 systems (*C. trachomatis/N. gonorrhoeae*, HPV, *T. vaginalis/M. genitalium*) 	Qualitative	96	3.5 hours	Urine, vaginal swab, endocervical swab	DNA	CE-IVD FDA	PCR

Table 10 (continued): Molecular laboratory-based platforms for the detection of *Neisseria gonorrhoeae*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Seegene Allplex, Anyplex and Seeplex test kits; open system (*C. trachomatis*/*N. gonorrhoeae*, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*/*M. genitalium*, HSV, HPV) 	Quantitative	N/A	N/A	N/A	DNA	CE-IVD	Real-time PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: U.S. Food and Drug Administration; HPA: hybridization protection assay; N/A: not applicable; PCR: polymerase chain reaction; qPCR: quantitative PCR; rPCR: random PCR; rRNA: ribosomal RNA; RT–PCR: PCR with reverse transcription; SDA: strand displacement amplification; TMA: transcription-mediated amplification.

Note: The information in this table is specific for *Neisseria gonorrhoeae* although most assays detect other STIs; see **Table 22** in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Table 11: Molecular platforms for use at POC or near-POC for *Neisseria gonorrhoeae*

Manufacturer/test (device)	Reporting	TAT	Sample type	Approval status	Test technology
AmplexDiagnostics eazyplex Complete STDs (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*), eazyplex STD *C. trachomatis*/*N. gonorrhoeae* (Genie III platform) 	Qualitative	N/A	Urethral, rectal/anal, vaginal, cervical and pharyngeal swab	CE-IVD	N/A
binx io *C. trachomatis*/*N. gonorrhoeae* assay (*io* system)  	Qualitative	30 minutes	Vaginal swab, male urine	FDA CE-IVD CLIA-waived	N/A
Bosch Healthcare Solutions STI Multiplex Array/ Vivalytic Analyzer (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV) 	Qualitative	2 hours 30 minutes	Urogenital swab, urine specimen	CE-IVD	Biochip array

Table 11 (continued): Molecular platforms for use at POC or near-POC for *Neisseria gonorrhoeae*

Manufacturer/test (device)	Reporting	TAT	Sample type	Approval status	Test technology
Cepheid Xpert *C. trachomatis/N. gonorrhoeae* assay (GeneXpert System) 	Qualitative	90 minutes	Urine, endocervical swab, rectal swab, throat swab, vaginal swab	FDA CE-IVD	N/A
HiberGene Diagnostics HG *C. trachomatis/N. gonorrhoeae* Combo test (HG Swift and HG Swift Plus) 	Qualitative	<60 minutes for positive, 70 minutes for negative	Vaginal swab, urine	CE-IVD	LAMP
Molbio Diagnostics Truenat *C. trachomatis/N. gonorrhoeae* assay (Truelab Real Time microPCR System) 	Semiquantitative	35 minutes	Endocervical swab, vaginal swab, male urethral swab, urine	CE-IVD	microPCR
Molbio Diagnostics Truenat *N. gonorrhoeae* assay (Truelab Real Time micro PCR System) 	Semiquantitative	35 minutes	Endocervical swab, vaginal swab, male urethral swab, urine	CE-IVD	microPCR
Ustar Biotechnologies *C. trachomatis/N. gonorrhoeae* (CPA assay) (EasyNAT Platform) 	Qualitative	50 minutes	Male urinary tract swab, cervical swab	N/A	CPA
Ustar Biotechnologies *N. gonorrhoeae* DNA (CPA assay) (EasyNAT platform) 	Qualitative	50 minutes	Male urinary tract swab, cervical swab	CE-IVD	CPA

Table 11 (continued): Molecular platforms for use at POC or near-POC for *Neisseria gonorrhoeae*

Manufacturer/test (device)	Reporting	TAT	Sample type	Approval status	Test technology
Visby Medical *C. trachomatis/N. gonorrhoeae/T. vaginalis* test 	Qualitative	50 minutes	Vaginal swab	FDA CLIA-waived	RT–PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CLIA: Clinical Laboratory Improvement Amendments waiver; CPA: cross priming amplification; FDA: U.S. Food and Drug Administration; LAMP: loop-mediated isothermal amplification; N/A: not applicable; RT–PCR: polymerase chain reaction with reverse transcription.

Note: The information in this table is specific for *N. gonorrhoeae* although most assays detect other STIs; see **Table 23** in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Detection of *Trichomonas vaginalis*: culture system and lateral flow

Diagnosis of *T. vaginalis* infection has traditionally been performed by microscopy of genital secretions, requiring immediate specimen preparation and reading of results with low sensitivity. There are currently two commercially available non-molecular tests to detect *T. vaginalis*. The BioMed Diagnostics InPouch *T. vaginalis* is an innovative self-contained culture system that allows for specimen storage and transport and is user-friendly. The Sekisui Diagnostics OSOM Trichomonas Test is an antigen detection RDT. Both are briefly described here.

InPouch TV (BioMed Diagnostics): The InPouch TV system is a commercially available plastic envelope method for *T. vaginalis* culture. It is a self-contained broth medium device for the recovery and detection of *T. vaginalis* from female vaginal samples or male urethral and urine samples. The InPouch TV is designed to be user-friendly and convenient and allows for early microscopic detection by culture confirmation of *T. vaginalis*. The pouch consists of a high-barrier, oxygen-resistant plastic with two V-shaped chambers connected by a narrow passage. Together, these allow the user to easily inoculate a specimen, immediately observe the specimen by wet mount and store and transport it before transfer to the laboratory for incubation and recording.

OSOM Trichomonas Test (Sekisui Diagnostics): The OSOM Trichomonas Test is an antigen detection-based RDT. Studies showed that it performed reasonably well when compared to wet mount and culture *(43, 44)*. The test has a turnaround time of 10 minutes.

Molecular tests for *Trichomonas vaginalis*

Like testing for *C. trachomatis* and *N. gonorrhoeae*, there are several reliable molecular laboratory platforms for *T. vaginalis* testing that are described in **Table 12**. In addition, POC and near-POC platforms can also detect *T. vaginalis* (see **Table 13**).

Table 12: Molecular laboratory-based platforms for the detection of *Trichomonas vaginalis*

Manufacturer/test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Abbott Alinity m System (*C. trachomatis/N. gonorrhoeae/T. vaginalis/M. genitalium*, high-risk HPV) 	Qualitative	N/A	Less than 115 minutes	Endocervical swab, vaginal swab, urine, oropharyngeal	RNA	CE-IVD	RT–PCR

Table 12 (continued): Molecular laboratory-based platforms for the detection of *Trichomonas vaginalis*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
BD Viper XTR system (ProbeTec *C. trachomatis* Q^x, gonorrhoea Q^x, *T. vaginalis* Q^x, HSV-1 and two Q^x Amplified DNA assays) 	Qualitative	96 with continuous loading	40 minutes	Endocervical swab, vaginal swab, female urine specimen	DNA	FDA CE-IVD	SDA
BD MAX system (*C. trachomatis*/ gonococcus/*T. vaginalis* assay) 	Qualitative	24	2.5–3 hours	Male urine (*C. trachomatis*, gonorrhoea), female urine (*C. trachomatis*, gonorrhoea, *T. vaginalis*), endocervical swab (*C. trachomatis*, gonorrhoea, *T. vaginalis*), vaginal swab (*C. trachomatis*, gonorrhoea, *T. vaginalis*)	DNA	FDA	Real-time PCR
Bioneer ExiStation Universal Molecular Diagnostic System (AccuPower assays – *C. trachomatis*, *C. trachomatis* and *N. gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*/ *M. genitalium*, *T. vaginalis*/ HSV, HPV), with ExiPrep and Exicycler 	N/A	N/A	N/A	Urine, vaginal swab	DNA	CE-IVD	Real-time PCR

Table 12 (continued): Molecular laboratory-based platforms for the detection of *Trichomonas vaginalis*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Hologic Panther System (Aptima assays – Combo 2 *C. trachomatis*/*N. gonorrhoeae*, herpes simplex viruses 1 and 2, HPV 16 18/45 genotype, HPV, *M. genitalium*, *T. vaginalis*) 	Qualitative	120	3 hours (first result), 5 minutes each thereafter	Endocervical swab, vaginal swab, specimen in preservCyt solution	rRNA	CE-IVD FDA	TMA, HPA
QIAGEN N.V. NeuMoDx assays (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*), with NeuMoDx 288 and 96 	Qualitative	N/A	N/A	Urine	DNA	CE-IVD	Real-time PCR
Roche cobas 6800/8800 systems (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*) 	Qualitative	96	3.5 hours	Urine, vaginal swab, endocervical swab, cervical specimen	DNA	CE-IVD FDA	PCR
Seegene Allplex, Anyplex and Seeplex test kits; open system (*C. trachomatis*/*N. gonorrhoeae*, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*/*M. genitalium*, HSV, HPV) 	Quantitative	N/A	N/A	N/A	DNA	CE-IVD	Real-time PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: Food and Drug Administration; HPA: hybridization protection assay; N/A: not applicable; PCR: polymerase chain reaction; RT–PCR: PCR with reverse transcription; SDA: strand displacement amplification; TMA: transcription-mediated amplification.

Note: The information in this table is specific for *Trichomonas vaginalis* although most assays detect other STIs; see **Table 22** in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Table 13: Molecular platforms for use at POC or near-POC for the detection of *Trichomonas vaginalis*

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
Bosch Healthcare Solutions STI Multiplex Array/ Vivalytic Analyzer (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV) 	Qualitative	2 hours 30 minutes	Urogenital swab, urine specimen	CE-IVD	Biochip array
Cepheid Xpert TV assay (GeneXpert System) 	Qualitative	1 hour	Male and female specimens	FDA CE-IVD	N/A
Molbio Diagnostics Truenat TV assay (Truelab Real Time microPCR System)  	Semi-quantitative	35 minutes	Endocervical swab, vaginal swab, male urethral swab, urine	CE-IVD	microPCR
QuidelOrtho SOLANA Trichomonas Assay 	N/A	50 minutes	Vaginal swab, urine	N/A	HDA
Ustar Biotechnologies TV DNA (CPA Assay) (EasyNAT Platform)  	Qualitative	50 minutes	Urine, urogenital swab	CE-IVD	CPA
Visby Medical *C. trachomatis*/*N. gonorrhoeae*/*T. vaginalis* test 	Qualitative	50 minutes	Vaginal swab	FDA CLIA-waived	RT–PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CLIA: Clinical Laboratory Improvement Amendments waiver; CPA: cross priming amplification; FDA: U.S. Food and Drug Administration; HDA: helicase-dependent amplification; N/A: not applicable; RT–PCR, polymerase chain reaction with reverse transcription.

Note: The information in this table is specific for *Trichomonas vaginalis* although most assays detect other STIs; see **Table 23** in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Molecular tests for *Mycoplasma genitalium*

Molecular testing has now become the gold standard for detecting *M. genitalium* (see **Table 14**). Both PCR-based and TMA-based molecular tests are available for *M. genitalium*. In addition, POC and near-POC platforms are available to detect *M. genitalium* (shown in **Table 15**).

Table 14: Molecular laboratory-based platforms for the detection of *Mycoplasma genitalium*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Abbott Alinity m System (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, high-risk HPV) 	Qualitative	N/A	Less than 115 minutes	Endocervical swab, vaginal swab, urine, oropharyngeal	RNA	CE-IVD	RT–PCR
Bioneer ExiStation Universal Molecular Diagnostic System (AccuPower assa–s - *C. trachomatis*, *C. trachomatis* and *N. gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*/*M. genitalium*, *T. vaginalis*, *T. vaginalis*/ HSV, HPV), with ExiPrep and Exicycler 	N/A	N/A	N/A	Urine, vaginal swab	DNA	CE-IVD	Real-time PCR
ELITechGroup Solutions STI ELITe MGB panel (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*) 	N/A	N/A	2.5 hours	Urine, cervical swab	DNA	CE-IVD	Real-time PCR

Table 14 (continued): Molecular laboratory-based platforms for the detection of *Mycoplasma genitalium*

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Hologic Panther System (Aptima assays – Combo 2 *C. trachomatis*/*N. gonorrhoeae*, herpes simplex viruses 1 and 2, HPV 16 18/45 genotype, HPV, *M. genitalium*, *T. vaginalis*) 	Qualitative	120	3 hours (first result), 5 minutes each thereafter	Endocervical swab, Vaginal swab, male urethral swab, Penile meatal swab, urine	rRNA	CE-IVD FDA	TMA, HPA
QIAGEN N.V. NeuMoDx assays (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*), with NeuMoDx 288 and NeuMoDx 96 	Qualitative	N/A	N/A	Urine	DNA	CE-IVD	Real-time PCR
Roche cobas 6800/8800 systems (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*) 	Qualitative	96	3.5 hours	Meatal swab	DNA	CE-IVD FDA	PCR
Seegene Allplex, Anyplex and Seeplex test kits; open system (*C. trachomatis*/*N. gonorrhoeae*, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV, HPV) 	Quantitative	N/A	N/A	N/A	DNA	CE-IVD	Real-time PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: U.S. Food and Drug Administration; HPA: hybridization protection assay; N/A: not applicable; PCR: polymerase chain reaction; rRNA: ribosomal RNA; RT–PCR: PCR with reverse transcription; TMA: transcription-mediated amplification.

Note: The information in this table is specific for *M. genitalium*, although most assays detect other STIs; see **Table 22** in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Table 15: Molecular platforms for use at POC or near-POC for the detection of *M. genitalium*

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
AmplexDiagnostics eazyplex STD complete (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*) (Genie III platform) 	Qualitative	N/A	Urethral, rectal/anal, vaginal, cervical, and pharyngeal swab	CE-IVD	N/A
Bosch Healthcare Solutions STI Multiplex Array/ Vivalytic Analyzer (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV) 	Qualitative	2 hours 30 minutes	Urogenital swab, urine specimen	CE-IVD	Biochip array
HiberGene Diagnostics *M. genitalium* test 	Qualitative	<60 minutes for positive, 70 minutes for negative	Vaginal swab, urine	CE-IVD	LAMP
Ustar Biotechnologies *M. genitalium* DNA (CPA assay) (EasyNAT Platform)  	Qualitative	50 minutes	Urine	CE-IVD	CPA

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CPA: cross priming amplification; LAMP: loop-mediated isothermal amplification; N/A: not applicable.

Note: The information in this table is specific for *M. genitalium* although most assays detect other STIs; see Table 23 in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Lateral flow tests for herpes simplex virus type 1 (HSV-1) and type 2 (HSV-2)

HSV type-specific antibody tests are based on the detection of HSV-specific glycoprotein G1 (gG1) (HSV-1) and glycoprotein G2 (gG2) (HSV-2) using native, purified or recombinant gG1 or Gg2 as antigens (3, 30). To assure accuracy, gG-based serological assays should be used *(3)*. Laboratory-based serological assays are commercially available. Assays that can identify both HSV-1 and HSV-2 are described in **Table 16**.

Table 16: Herpes simplex virus type-specific lateral flow laboratory tests

Manufacturer/test	Reporting	Purpose	Detection	Intended subjects	Sample type	Approval stage	Test technology
Bio-Rad Laboratories BioPlex 2200 HSV-1 and HSV-2 kit	Qualitative	Detection and differentiation	IgG antibodies	Sexually active adults, expectant mothers	Serum, EDTA plasma, heparinized plasma	FDA	MFIA
DiaSorin LIAISON HSV-1 Type Specific IgG, LIAISON HSV-2 IgG and HSV-1/2 IgG Assays	Qualitative	Detection	Type-specific IgG antibodies	Sexually active adults, expectant mothers	Serum	FDA	Chemiluminescence
Focus Diagnostics (DiaSorin Group) HerpeSelect 1 and 2 ELISA kit	Qualitative	Detection	IgG antibodies	Sexually active adults, pregnant women	Serum	FDA	ELISA
Focus Diagnostics (DiaSorin Group) HerpeSelect 1 and 2 Immunoblot Assay	Qualitative	Detection and differentiation	IgG antibodies	Sexually active adults, pregnant women	Serum	FDA	
Roche Elecsys HSV-1 IgG and HSV-2 IgG Assays	Qualitative	Determination	IgG antibodies	Sexually active adults, pregnant women	Serum, lithium-heparin plasma, K2-EDTA plasma, K3-EDTA plasma	FDA	ECLIA
Siemens ADVIA Centaur HSV1 and HSV2 Assays	Qualitative	Determination	IgG antibodies	Sexually active adults, expectant women	Serum and plasma (EDTA and lithium-heparin)	FDA	CIA
ZEUS Scientific ELISA HSV gG-2 IgG and ELISA HSV gG-1 IgG Test Systems	Qualitative	Detection	Type-specific IgG antibodies	Sexually active adults, pregnant women	Serum	CE-IVD FDA	ELISA

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CIA: confocal immunoanalysis; ECLIA: electrochemiluminescence immunoassay; EDTA: ethylenediaminetetraacetic acid; ELISA: enzyme-linked immunosorbent assay; FDA: U.S. Food and Drug Administration; IgG: immunoglobulin G; MFIA: multiplexed flow immunoassay.

Test descriptions for the HSV lateral flow tests are shown below.

BioPlex 2200 HSV-1 and HSV-2 Kit (Bio-Rad Laboratories): The BioPlex 2200 HSV-1 and HSV-2 Kit from Bio-Rad Laboratories is a multiplex flow immunoassay for the qualitative detection and differentiation of IgG antibodies to HSV-1 and HSV-2 in human serum and EDTA or heparinized plasma. The test is intended for use in sexually active adults and expectant mothers as an aid for the presumptive diagnosis of HSV-1 and HSV-2 infection. The test is intended for use with the Bio-Rad BioPlex 2200 System, which is described in detail earlier in this report.

LIAISON HSV-1 Type Specific IgG, LIAISON HSV-2 IgG and HSV-1/2 IgG Assays (DiaSorin): DiaSorin offers several HSV tests as part of its TORCH line of assays. The assays are for use on its LIASON family of analysers, which are described in this report in connection with the LIAISON *Treponema* Assay.

Like its other immunoassays, the LIAISON HSV-1 Type Specific IgG, HSV-2 IgG and HSV-1/2 IgG Assays use CLIA technology for the qualitative detection of type-specific IgG antibodies to HSV-1 or HSV-2 in human serum. The assays are indicated for testing sexually active adults and expectant mothers. Recombinant antigens specific for IgG to HSV-1 or HSV-2 are used to coat the magnetic particles and are used in the tracer when linked to an isoluminol derivative (isoluminol–antigen conjugate). During the first incubation step, antibodies present in the calibrators, samples or controls bind to the solid phase.

During the second incubation, the antibody conjugate reacts with the antibodies already bound to the solid phase. After each incubation, the unbound material is removed with a wash cycle. Subsequently, starter reagents are added and a flash chemiluminescence reaction is induced. The light signal, and hence the amount of isoluminol–antigen conjugate, is measured by a photomultiplier as RLUs and is indicative of total antibodies to HSV-1 or HSV-2 present in calibrators, controls or samples.

HerpeSelect 1 and 2 ELISA Kit (Focus Diagnostics, part of the DiaSorin Group): In addition to its HSV-1 and HSV-2 immunoblot assays, Focus Diagnostics also makes two ELISA assays, the HerpeSelect 1 ELISA IgG assay and the HerpeSelect 2 ELISA IgG assay. These assays are intended for the qualitative detection of the presence or absence of human IgG class antibodies to HSV-1 or HSV-2 in human sera, depending on the test, and may be performed manually or on automated ELISA systems (e.g. ELISA systems from Thermo Fisher Scientific, EUROIMMUN and Dynex, among others). The tests are intended for testing sexually active adults or pregnant women to aid the presumptive diagnosis of HSV infection.

Regarding either the HerpeSelect 1 or 2 ELISA IgG assay, the polystyrene microwells are coated with recombinant gG1 or gG2 antigen, depending on the test. Diluted serum samples and controls are incubated in the wells to allow specific antibodies present in the samples to react with the antigen. Nonspecific reactants are removed by washing, and POD-conjugated anti-human IgG is added and reacts with specific IgG. Excess conjugate is removed by washing. Enzyme substrate and chromogen are added, and the colour is allowed to develop. After adding a stop reagent, the resultant colour change is quantified by a spectrophotometric reading of optical density (OD). The sample OD readings are compared with a reference cut-off OD reading to determine results.

HerpeSelect 1 and 2 Immunoblot Assay (Focus Diagnostics, part of the DiaSorin Group): This test is intended for the qualitative detection and differentiation of human IgG class antibodies to HSV-1 and HSV-2 in human sera in sexually active adults or pregnant women as an aid in the presumptive diagnosis of HSV-1 and HSV-2 infection.

The HerpeSelect 1 and 2 Immunoblot is a manual test that purifies native and recombinant antigens and applies them onto a nitrocellulose membrane, which is dried and cut into strips. The test is a two-stage procedure. In the first stage, patient serum is diluted and incubated with individual antigen strips. If antibodies to HSV type-specific antigens or HSV common antigens are present in the serum, the antibodies bind to the antigens immobilized on the nitrocellulose membranes. In the second stage, the blots are incubated with ALP-conjugated goat anti-human IgG, substrate is added and a coloured precipitate forms where the anti-human conjugate has bound. The coloured band reactivity is then interpreted.

Elecsys HSV-1 IgG and HSV-2 IgG Assays (Roche Diagnostics): The Elecsys HSV-1 IgG and HSV-2 IgG Assays from Roche are for the qualitative determination of IgG class antibodies to HSV-1 or HSV-2, depending on the test, in human serum and lithium-heparin plasma, K2-EDTA plasma and K3-EDTA plasma. The tests are intended for sexually active adults and pregnant women as an aid in the presumptive diagnosis of HSV-1 or HSV-2 infection, as the case may be. The assays use ECLIA technology and are intended for use on the family of cobas e analysers, which are described in detail earlier in this report.

ADVIA Centaur HSV1 and HSV2 Assays (Siemens): The ADVIA Centaur Herpes-1 IgG (HSV-1) and Herpes-2 IgG (HSV-2) assays from Siemens are in vitro diagnostics used in the qualitative determination of IgG antibodies to HSV-1 or HSV-2, depending on the test, in human serum and plasma (EDTA and lithium-heparin). The tests are intended for sexually active adults or expectant women as an aid in the presumptive diagnosis of HSV infections. The tests are designed for use on the automated ADVIA Centaur systems, which are described in detail earlier in this report.

ZEUS ELISA HSV gG-2 IgG Test System and ZEUS ELISA HSV gG-1 IgG Test System (ZEUS Scientific): The two ZEUS ELISA HSV tests are intended for the qualitative detection of type-specific IgG class antibodies to HSV-1 or HSV-2 (depending on the assay) in human serum. They are intended for testing sexually active individuals or pregnant women for aiding in the presumptive diagnosis of either HSV-1 or HSV-2 infection.

To perform the assays, the wells of plastic microwell strips are sensitized by passive adsorption with recombinant HSV-1 antigen or with inactivated affinity purified HSV-2 antigen, depending on the test. In either case, the test procedure involves three steps: *(1)* properly diluted test sera are incubated at 20–25 °C for approximately 25 minutes in antigen-coated microwells, after which the plate is washed to remove unbound antibody; *(2)* POD-conjugated goat anti-human IgG is added to the wells and the plate is again incubated at 20–25 °C for approximately 25 minutes whereupon the conjugate will react with IgG antibody immobilized on the solid phase; and *(3)* after additional washes, to remove unreacted conjugate, the microwells containing the immobilized POD conjugate are incubated with POD substrate solution, which produces a colour change; after 10–15 minutes, the reaction is stopped and the colour intensity of the solution is measured photometrically within 30 minutes. These assays are primarily manual, although microwell automated wash systems can be used. In addition to type-specific serological assays, molecular testing is adaptable for typing.

Molecular tests for herpes simplex virus type 1 (HSV-1) and type 2 (HSV-2)

Molecular testing has been shown to be more sensitive, compared to viral cell culture, and is generally the test of choice (3, 15). There are several commercially available molecular laboratory-based systems that can detect HSV-1 and HSV-2 and these are described in **Table 17**. In addition, there are POC and near-POC molecular platforms that have the capability to perform HSV testing; these are described in **Table 18**.

Table 17: Molecular laboratory-based platforms for detection of herpes simplex virus (HSV)

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
BD Viper XTR system (ProbeTec *C. trachomatis* Q^x, gonococcus Q^x, *T. vaginalis* Q^x, HSV-1 and 2 Q^x Amplified DNA assays) 	Qualitative	96 with continuous loading	40 minutes	Anogenital lesion	DNA	FDA	SDA
Bioneer ExiStation Universal Molecular Diagnostic System (AccuPower assays – *C. trachomatis*, *C. trachomatis* and *N. gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*/*M. genitalium*, *T. vaginalis*, *T. vaginalis*/ HSV, HPV), with ExiPrep and Exicycler 	N/A	N/A	N/A	Urine, vaginal swab	DNA	CE-IVD	Real-time PCR

Table 17 (continued): Molecular laboratory-based platforms for detection of herpes simplex virus (HSV)

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Hologic Panther System (Aptima assays – Combo 2 *C. trachomatis/N. gonorrhoeae*, herpes simplex viruses 1 & 2, HPV 16 18/45 genotype, HPV, *M. genitalium*, *T. vaginalis*) 	Qualitative	120	3 hours (first result), 5 minutes each thereafter	Anogenital skin lesion	mRNA	CE-IVD FDA	TMA, HPA
Meridian Bioscience Alethia (HSV 1 & 2 assay) 	Qualitative	1–10	<1 hour	Cutaneous/ mucocutaneous lesion specimen	DNA	FDA	LAMP
QIAGEN N.V. QIAsymphony artus kits (*C. trachomatis/N. gonorrhoeae* QS- Rotor-Gene Q, HSV-1/2 Rotor-Gene Q), with SP/AS and Rotor-Gene Thermocyclers 	Qualitative	96	N/A	Plasma	DNA	CE-IVD	Real-time PCR
Roche cobas 4800 system (*C. trachomatis/N. gonorrhoeae* version 2.0, HSV-1 and HSV-2, HPV) 	Qualitative	96	30 minutes	External anogenital lesion	DNA	CE-IVD FDA	Real-time PCR

Table 17 (continued): Molecular laboratory-based platforms for detection of herpes simplex virus (HSV)

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Seegene Allplex, Anyplex and Seeplex test kits; open system (*C. trachomatis*/*N. gonorrhoeae*, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis* and *M. genitalium*, HSV, HPV) 	Quantitative	N/A	N/A	N/A	DNA	CE-IVD	Real-time PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: U.S. Food and Drug Administration; HPA: hybridization protection assay; IgG: immunoglobulin G; IgM: immunoglobulin M; mRNA: messenger RA; N/A: not applicable; PCR: polymerase chain reaction; SDA: strand displacement amplification; TMA: transcription-mediated amplification.

Note: The information in this table is specific for HSV although most assays detect other STIs; see Table 22 in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Table 18 : Molecular platforms for use at POC and near-POC for the detection of HSV

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
Bosch Healthcare Solutions STI Multiplex Array/ Vivalytic Analyzer (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV) 	Qualitative	2 hours 30 minutes	Urogenital swab, urine specimen	CE-IVD	Biochip array
HiberGene Diagnostics HSV test 	Qualitative	<40 minutes for positive, 50 minutes for negative	Urogenital swab	CE-IVD	LAMP
Luminex ARIES HSV 1 and HSV2 Assay (ARIES and ARIES M1 Systems) 	Qualitative	2 hours	Cutaneous or mucocutaneous lesion	FDA CE-IVD	Real-time PCR

Table 18 (continued): Molecular platforms for use at POC and near-POC for the detection of HSV

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
QuidelOrtho SOLANA HSV1 + 2/varicella zoster virus assay 	N/A	50 minutes	Vaginal swab, urine	N/A	HDA
QuidelOrtho Lyra Direct HSV 1+2/ varicella zoster virus assay	N/A	Less than 70 minutes	Cutaneous or mucocutaneous swab	N/A	PCR
QuidelOrtho AmpliVue HSV 1+2 Assay 	Qualitative	60 minutes	Cutaneous or mucocutaneous swab	N/A	Lateral flow strip
Ustar Biotechnologies EasyNAT HSV 1 & 2 Assay  	Qualitative	50 minutes	Urogenital swab	CE-IVD	CPA

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CPA: cross priming amplification; FDA: U.S. Food and Drug Administration; HDA: helicase-dependent amplification; LAMP: loop-mediated isothermal amplification; N/A: not applicable; PCR: polymerase chain reaction.

Note: The information in this table is specific for HSV; see Table 23 in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Lateral flow test for human papillomavirus

There is only one commercially available lateral flow assay, which is shown in **Table 19**. Kelly et al. concluded that the low sensitivity, but higher specificity, of the OncoE6 assay for CIN2+ detection suggests that it might be "useful as a 'screen-and-treat' or triage test", but further studies are needed *(45)*.

Table 19. Lateral flow test for human papillomavirus

Manufacturer/ Test	Throughput	Turnaround time	Sample type	Detection	Approval stage	Test technology
Arbor Vita Corporation Onco*E6* test 	45 specimens per operator, per day	2–2.5 hours	Pap smear, liquid ThinPrep	E6 oncoprotein	N/A	Lateral flow immunoassay

N/A: not applicable.

OncoE6 Assay (Arbor Vita Corporation): The Onco*E6* test from Arbor Vita Corporation is a lateral flow immunoassay that detects the *E6* oncoprotein from two high-risk HPV types (HPV-16, HPV-18), which cause approximately 75% of invasive cervical cancer (ICC). The Onco*E6* test is in a dipstick-like format; it is simple, quick, non-invasive and does not require refrigeration. The test is compatible with specimens collected for either a regular Pap smear or liquid ThinPrep from Arbor Vita.

The Onco*E6* does not require complex equipment for processing and can process 45 specimens per operator per day, a volume that can be processed in a clinic within 2–2.5 hours.

Molecular tests for human papillomavirus

In 2021, WHO recommended molecular-based testing as first-line screening for HPV. Currently available molecular tests for the detection of the human papillomavirus are described in **Table 20** and elsewhere *(46)*. There are also several POC or near-POC platform and assay options currently available for use in resource-limited settings as shown in **Table 21**.

Table 20: Molecular laboratory-based platforms for detection of human papillomavirus (HPV)

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Abbott Alinity m System (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, high-risk HPV) 	Qualitative	N/A	Less than 115 minutes	Cervical specimen	DNA	CE-IVD	RT–PCR
Abbott RealTime *m*2000 system (high-risk HPV assay) 	Qualitative	96 samples per run	N/A	Asymptomatic: vaginal swab, urine swab	DNA	CE-IVD	Real-time PCR
BD Viper LT system (ProbeTec *C. trachomatis* Q^{x}, gonococcus Q^{x} assays and Onclarity HPV) 	Qualitative	30	3.5 hours	Cervical specimen	DNA	FDA CE-IVD	SDA

Table 20 (continued): Molecular laboratory-based platforms for detection of human papillomavirus (HPV)

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Bioneer ExiStation Universal Molecular Diagnostic System (AccuPower assays – *C. trachomatis*, *C. trachomatis* and *N. gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*/*M. genitalium*, *T. vaginalis*, *T. vaginalis*/ HSV, HPV), with ExiPrep and Exicycler 	N/A	N/A	N/A	Cervical swab, liquid-based cytology specimen	DNA	CE-IVD	Real-time PCR
Hologic Panther System (Aptima assays – Combo 2 *C. trachomatis*/*N. gonorrhoeae*, herpes simplex viruses 1 & 2, HPV 16 18/45 genotype, HPV, *M. genitalium*, *T. vaginalis*) 	Qualitative	120	3 hours (first result), 5 minutes each thereafter	Cervical specimen and liquid cytology specimens	mRNA	CE-IVD FDA	TMA, HPA
QIAGEN N.V. NeuMoDx assays (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*), with NeuMoDx 288 and NeuMoDx 96 	Qualitative	N/A	N/A	Cervical specimen	DNA	CE-IVD	Real-time PCR

Table 20 (continued): Molecular laboratory-based platforms for detection of human papillomavirus (HPV)

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Roche cobas 4800 system (CT/NG version 2.0, HSV 1 and 2, HPV) 	Qualitative	96	30 minutes	Cervical specimen	DNA	CE-IVD FDA	Real-time PCR
Roche cobas 6800/8800 systems (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*) 	Qualitative	96	3.5 hours	Cervical specimen	DNA	CE-IVD FDA	PCR
Seegene Allplex, Anyplex and Seeplex test kits; open system (*C. trachomatis*/*N. gonorrhoeae*, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis* and *M. genitalium*, HSV, HPV) 	Quantitative	N/A	N/A	N/A	DNA	CE-IVD	Real-time PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: U.S. Food and Drug Administration; HPA: hybridization protection assay; IgG: immunoglobulin G; IgM: immunoglobulin M; MFIA: multiplexed flow immunoassay; N/A: not applicable; PCR: polymerase chain reaction; SDA: strand displacement amplification; TMA: transcription-mediated amplification.

Note: The information in this table is specific for HPV although most assays detect other STIs; see **Table 22** in the section "Molecular multiplex laboratory-based platform systems for detection of STIs".

Table 21: Molecular platforms for use at POC or near-POC for the detection of human papillomavirus (HPV)

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
Cepheid Xpert HPV assay (GeneXpert System) 	Qualitative	<1 hour	Endocervical sample	CE-IVD	N/A
Molbio Diagnostics Truenat HPV-HR assay (Truelab Real Time microPCR System)  	Semi-quantitative	1 hour	Cervical specimen	CE-IVD	microPCR
Ustar Biotechnologies EasyNAT HPV 6/11 assay  	Qualitative	50 minutes	Endocervical swab, urogenital swab	CE-IVD	CPA

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CPA: cross priming amplification; N/A: not applicable.

careHPV System (QIAGEN): The *care*HPV Test (QIAGEN), which has been added to the WHO list of prequalified IVDs, provides primary, stand-alone screening for high-risk HPV in women aged 30 years and older. The test, which is a nucleic acid hybridization assay with signal amplification using microplate chemiluminescence, detects 14 high-risk HPV types in about 2.5 hours, which permits same-day follow-up. A multicentre clinical study conducted to validate the *care*HPV Test reported a clinical sensitivity of 88% and clinical specificity of 85% for CIN2+, although results may vary depending on the specific patient population, including slightly lower sensitivity and specificity in women living with HIV and AIDS *(47)*.

The *care*HPV System includes the *care*HPV Test Controller, *care*HPV Test Shaker, *care*HPV Test Luminometer and the *care*HPV Test Magnetic Plate Holder. The automated components are designed for a universal power supply and operate on AC mains electricity or a lead–acid battery. QIAGEN also provides simple sample collection materials with multiple day stability in the form of the *care*HPV Sample Collection Device, which consists of the *care*Brush and *care*HPV Collection Medium. These materials permit both health care provider sampling and self-sampling, which has the possibility to increase uptake and screening.

Molecular multiplex tests for detecting *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *Trichomonas vaginalis*, *Mycoplasma genitalium*, herpes simplex viruses 1 and 2, and human papillomavirus

Below are the commercially available IVD molecular systems previously described under of each of the STI infections that are the subject of this report. The systems include an automated platform compatible with assays able to detect multiple STIs. Many systems have separate automated instruments to perform sample preparation and nucleic acid extraction.

Table 22 describes laboratory-based platforms and **Table 23** describes platforms that could be used at POC or near-POC. More detailed descriptions for each platform and test are provided below the tables.

Table 22: Molecular multiplex laboratory-based platform systems for detection of sexually transmitted infections

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Abbott Alinity m System (*C. trachomatis*/*N. gonorrhoeae*/*T. vaginalis*/*M. genitalium*, high-risk HPV) 	Qualitative	N/A	Less than 115 minutes	Endocervical swab, vaginal swab, urine, oropharyngeal; rectal (*C. trachomatis* and *N. gonorrhoeae*); cervical specimen (HPV)	RNA (*C. trachomatis*, *T. vaginalis*, *M. genitalium*); DNA (*N. gonorrhoeae*, HPV)	CE-IVD	RT–PCR
Abbott RealTime m2000 system (*C. trachomatis*/*N. gonorrhoeae*, HPV) 	Qualitative	96 samples per run	N/A	Symptomatic: endocervical and vaginal swab, male urethral swab, urine swab (*C. trachomatis*/*N. gonorrhoeae*); asymptomatic: vaginal swab, urine swab (*C. trachomatis*/*N. gonorrhoeae*, HPV)	Genomic DNA (*N. gonorrhoeae*), plasma DNA (*C. trachomatis*); high-risk HPV DNA	FDA (*C. trachomatis*/*N. gonorrhoeae*) CE-IVD (HPV)	Real-time PCR
BD ProbeTec ET system (*C. trachomatis* / gonococcus) 	Qualitative	46 samples	3 hours	Endocervical swab, male urethral swab, urine	*C. trachomatis* or gonococcal DNA	FDA	SDA
BD Viper XTR system (ProbeTec *C. trachomatis* Q^x, GC Q^x, *T. vaginalis* Q^x, HSV-1 and HSV-2 Q^x Amplified DNA assays) 	Qualitative	96 with continuous loading	40 minutes	Endocervical swab, male urethral swab. Male and female urine specimens (*C. trachomatis*, gonococcus); anogenital lesion (HSV); endocervical swab, vaginal swab, female urine specimen (*T. vaginalis*)	DNA	FDA (gonococcus, *C. trachomatis*, *T. vaginalis*, HSV) CE-IVD (*T. vaginalis*, HSV)	SDA (gonococcus, *C. trachomatis*, *T. vaginalis*, HSV) rPCR (gonococcus, *C. trachomatis*,)
BD Viper LT system (ProbeTec CT Q^x, gonococcus Q^x assays, and Onclarity HPV) 	Qualitative	30	3.5 hours	Endocervical swab, male urethral swab, vaginal swab, female urine; cervical specimen (HPV)	DNA	FDA (gonococcus, *C. trachomatis*, HPV) CE-IVD (HPV)	SDA (gonococcus, *C. trachomatis*, HPV) rPCR (gonococcus, *C. trachomatis*)

Table 22 (continued): Molecular multiplex laboratory-based platform systems for detection of sexually transmitted infections

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
BD MAX system (*C. trachomatis*/ gonococcus/*T. vaginalis* assay) 	Qualitative	24	2.5–3 hours	Male urine (*C. trachomatis*, gonococcus), female urine (*C. trachomatis*, gonococcus, *T. vaginalis*), endocervical swab (*C. trachomatis*, gonococcus, *T. vaginalis*), vaginal swab (*C. trachomatis*, gonococcus, *T. vaginalis*)	DNA	FDA	Real-time PCR
Bioneer ExiStation Universal Molecular Diagnostic System (AccuPower assays – *C. trachomatis*, *C. trachomatis* and *N. gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*/*M. genitalium*, *T. vaginalis*, *T. vaginalis*/HSV, HPV), with ExiPrep and Exicycler 	N/A	N/A	N/A	Urine, vaginal swab (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*, *T. vaginalis*, HSV), urethral swab (*C. trachomatis*); cervical swab, liquid-based cytology specimen (HPV)	DNA	CE-IVD (except *C. trachomatis*/*N. gonorrhoeae*)	Real-time PCR
ELITechGroup Solutions STI ELITe MGB panel (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*) 	N/A	N/A	2.5 hours	Urine, cervical swab	DNA	CE-IVD	Real-time PCR
Hain Lifescience FluoroType system (*C. trachomatis*, *N. gonorrhoeae* assays), with GenoXtract and FluoroCycler PCR 	Quantita-tive	12	Varies by assay	Cervical swab, urine (*N. gonorrhoeae*, *C. trachomatis*); urethral swab (*N. gonorrhoeae*); ejaculate (*C. trachomatis*)	DNA	CE-IVD	qPCR

Table 22 (continued): Molecular multiplex laboratory-based platform systems for detection of sexually transmitted infections

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
Hologic Panther System (Aptima assays – Combo 2 *C. trachomatis*/*N. gonorrhoeae*, herpes simplex viruses 1 and 2, HPV 16 18/45 genotype, HPV, *M. genitalium*, *T. vaginalis* 	Qualitative	120	3 hours (first result), 5 minutes each thereafter	Endocervical swab (*C. trachomatis*/*N. gonorrhoeae*, *M. genitalium*, *T. vaginalis*); vaginal swab (*C. trachomatis*/*N. gonorrhoeae*, *M. genitalium*, *T. vaginalis*); gynaecological specimen (*C. trachomatis*/*N. gonorrhoeae*); cervical specimen and liquid cytology specimens (HPV); male urethral swab (*C. trachomatis*/*N. gonorrhoeae*, *M. genitalium*); penile meatal swab (*M. genitalium*); rectal swab, (*C. trachomatis*/*N. gonorrhoeae*); anogenital skin lesion (HSV); urine (*C. trachomatis*/*N. gonorrhoeae*, *M. genitalium*); throat swab, (*C. trachomatis*/*N. gonorrhoeae*); specimen in preservCyt solution (*T. vaginalis*)	rRNA (*C. trachomatis* or *N. gonorrhoeae*, *M. genitalium*, *T. vaginalis*); mRNA (HSV, HPV)	CE-IVD FDA	TMA, HPA
Meridian Bioscience Alethia (HSV 1 and 2 assay) 	Qualitative	1–10	<1 hour	Cutaneous/ mucocutaneous lesion specimen	DNA	FDA	LAMP
QIAGEN N.V. QIAsymphony artus kits (*C. trachomatis*/*N. gonorrhoeae* QS-Rotor-Gene Q, HSV-1/2 Rotor-Gene Q), with SP/ AS and Rotor-Gene Thermocyclers 	Qualitative	96	N/A	Vaginal swab, urine (*C. trachomatis*/*N. gonorrhoeae*); cerebrospinal fluid, plasma (HSV)	Plasmid (*C. trachomatis*), genomic DNA (*C. trachomatis*, *N. gonorrhoeae*), DNA (HSV)	CE-IVD	Real-time PCR

Table 22 (continued): Molecular multiplex laboratory-based platform systems for detection of sexually transmitted infections

Manufacturer/ test (device)	Reporting	Throughput	Turnaround time	Sample type	Detection	Approval status	Test technology
QIAGEN N.V. digene tests (HC2 *C. trachomatis*-gonococcus Dual ID, HC2 *C. trachomatis*-ID DNA, HC2 *C. trachomatis*/ gonococcus, HC2 gonococcus-ID, HPV) with Rapid Capture System 	Qualitative	352	In 8-hour shift	Cervical specimen	DNA	FDA	Chemilumi-nescence
QIAGEN N.V. NeuMoDx assays (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*/*M. genitalium*), with NeuMoDx 288 and NeuMoDx 96  	Qualitative	N/A	N/A	Urine (*C. trachomatis*/*N. gonorrhoeae*, *T. vaginalis*/*M. genitalium*), cervical specimen (HPV)	DNA	CE-IVD	Real-time PCR
Roche cobas 4800 system (*C. trachomatis*/*N. gonorrhoeae* version 2.0, HSV-1 and HSV-2, HPV) 	Qualitative	96	30 minutes	Vaginal swab, male urine (*C. trachomatis*, *N. gonorrhoeae*); external anogenital lesion (HSV); cervical specimen (HPV)	DNA	CE-IVD FDA	Real-time PCR
Roche cobas 6800/8800 systems (CT/NG, HPV, TV/ MG) 	Qualitative	96	3.5 hours	Urine, vaginal swab, endocervical swab (CT/NG, TV); cervical specimen (HPV, TV); meatal swab (MG)	DNA	CE-IVD FDA	PCR
Seegene Allplex, Anyplex and Seeplex test kits; open system (*C. trachomatis*/*N. gonorrhoeae*, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis* and *M. genitalium*, HSV, HPV) 	Quantita-tive	N/A	N/A	N/A	DNA	CE-IVD	Real-time PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: U.S. Food and Drug Administration; HPA: hybridization protection assay; LAMP: loop-mediated isothermal amplification; N/A: not applicable; PCR: polymerase chain reaction; qPCR: quantitative PCR; rPCR: random PCR; RT–PCR: PCR with reverse transcription; SDA: strand displacement amplification; TMA: transcription-mediated amplification.

Molecular multiplex laboratory-based platform systems for the detection of sexually transmitted infections

Alinity m System (Abbott): Abbott offers several STI assays designed for use on its Alinity m system. These assays are a combination 4-in-1 multiplex assay to detect and differentiate ***C. trachomatis***, ***N. gonorrhoeae, T. vaginalis*** and **M. genitalium**, as well as a **high-risk HPV**. The combination *C. trachomatis*/*N. gonorrhoeae*/*T. vaginalis*/*M. genitalium* test is an in vitro PCR with reverse transcription (RT–PCR) assay for the direct, qualitative detection and differentiation of RNA from *C. trachomatis*, DNA from *N. gonorrhoeae*, RNA from *T. vaginalis* and RNA from *M. genitalium*, to aid in the diagnosis of disease caused by infection from these microorganisms. The assay may be used with the following specimens from both symptomatic and asymptomatic individuals: endocervical swab specimens; clinician-collected vaginal swab specimens; self-collected vaginal swab specimens (in a clinical setting); female urine; and male urine. The assay may also be used to test oropharyngeal and rectal swab specimens from symptomatic and asymptomatic individuals for *C. trachomatis* and *N. gonorrhoeae*.

The Alinity m **high-risk HPV** assay is a qualitative in vitro test for the detection of DNA from 14 high-risk HPV genotypes (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68) in clinical specimens. The assay specifically identifies HPV genotypes 16, 18 and 45, while reporting the concurrent detection of the other high-risk genotypes (31, 33, 52, 58) and (35, 39, 51, 56, 59, 66, 68) at clinically relevant infection levels. The assay is intended for use *(1)* to screen patients with atypical squamous cells of undetermined significance (ASC-US) cervical cytology test results to determine the need for referral to colposcopy; *(2)* with cervical cytology to adjunctively screen to assess the presence or absence of the high-risk HPV genotype; *(3)* as a first-line primary screening test to identify women at increased risk for the development of cervical cancer or the presence of high-grade disease; and *(4)* to assess the presence or absence of HPV genotypes 16 and 18 to identify women at increased risk for the development of cervical cancer or the presence of high-grade disease with or without cervical cytology.

The Alinity m system is a continuous and random-access molecular analyser that uses RT–PCR technology. The system uses a single, universal, multi-collection device compatible with multiple specimens, which simplifies sample collection. It also eliminates batching procedures and provides a first result in less than 115 minutes.

m2000 system (Abbott): Abbott manufactures the Abbott RealTime ***C. trachomatis/N. gonorrhoeae*** assay, which is a real-time PCR assay for direct, qualitative detection of the genomic DNA of *N. gonorrhoeae* and the plasma DNA of *C. trachomatis* on its automated *m*2000 and *m* 24 systems. The assay may be used to test the following swabs from symptomatic patients: female endocervical swabs; clinician- or patient-collected vaginal swabs; male urethral swab specimens; and male and female urine swabs. The assay may also be used to test the following swabs from asymptomatic patients: clinician- or patient-collected vaginal swabs; and male and female urine swabs.

Abbott also offers the Abbott RealTime **high-risk HPV** assay, which is a qualitative in vitro molecular assay that uses homogeneous target amplification and detection technology for the detection of high-risk HPV DNA in cervical cells collected in liquid cytology medium. The assay is intended to detect 14 high-risk HPV genotypes—16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68—and to partially genotype 16 and 18 from the other 12 high-risk genotypes. The test may be used to detect high-risk HPV in cervical specimens from women with precancerous lesions of the cervix uteri (≥CIN2) and cervical cancer for cervical cancer screening.

The Abbott RealTime *C. trachomatis*/*N. gonorrhoeae* and high-risk HPV assays (and the other assays listed here) can be automated using the Abbott *m*2000*rt* for amplification and detection and one of three methods for sample preparation: *(1)* manual (for laboratories with low-throughput requirements); *(2)* the *m*24*sp* instrument (for laboratories with low-to-medium-throughput requirements); or *(3)* the m2000sp instrument (for laboratories with medium-to-high-throughput requirements).

The *m*24*sp* is a benchtop sample preparation and extraction device with a small footprint that is generally appropriate for facilities with medium-throughput requirements. It provides a variable extraction system (extraction output can be stored either in deep-well trays or 1.5-ml tubes) with ready-to-use and reusable reagents as well as flexible batch size capabilities.

The *m*2000*sp* is a larger and more automated sample preparation device than the *m*24*sp*. It is a high-throughput system with a maximum batch size of 96 samples per run; the reagents and tips required for extraction are loaded manually by the operator. When combined with the Abbott m2000rt, the amplification and detection instrument, the system can provide automation from barcoded laboratory tube through to patient result.

The Abbott *m*2000*rt* is the amplification and detection platform for use with manual extraction, the m24sp and the *m*2000*sp* instruments, as described earlier. It is a compact high-performance system weighing just over 34 kg and can run 96 samples at a time in about 3 hours of cycling time (not including time for sample preparation). The system will run both quantitative and qualitative analyses.

ProbeTec ET system (BD Diagnostics): BD Diagnostics offers BD ProbeTec ET amplified DNA assays, which allow direct and qualitative detection of ***C. trachomatis*** and **gonococcal** DNA in endocervical swabs, male urethral swabs, and male and female urine specimens (with and without preservative) that are collected from either symptomatic or asymptomatic patients.

The assay may be run on the BD ProbeTec ET system, which uses strand displacement amplification (SDA) technology as the amplification method and fluorescence energy transfer as the detection method to test for the presence of *C. trachomatis* and *N. gonorrhoeae* in the clinical specimens indicated. While the BD ProbeTec is relatively compact and requires no special room set-up, the system requires separate priming and amplification microwells, a pipettor, and a lysing rack and heater. Nonetheless, the system can generate *C. trachomatis*/*N. gonorrhoeae* results for up to 46 patient samples in 3 hours, or 276 *C. trachomatis*/*N. gonorrhoeae* results in one 8-hour shift. Total hands-on time is less than 2 minutes per sample.

BD Viper System with XTR technology, BD Viper LT system and BD COR PX/GX System (BD Diagnostics): BD offers the BD ProbeTec ***C. trachomatis*** Q^x, *N. gonorrhoeae* Q^x Amplified DNA assays for use on either the BD Viper System with XTR technology or the BD Viper LT system. Both systems use SDA and random PCR (rPCR) technology for the direct and qualitative detection of *N. gonorrhoeae* DNA or *C. trachomatis* DNA, respectively, in clinician-collected female endocervical and male urethral swab specimens, patient-collected vaginal swab specimens (in a clinical setting), and male and female urine specimens. The assays are indicated for use with asymptomatic and symptomatic females and symptomatic males to aid in the diagnosing of gonococcal urogenital disease. The assays are based on the simultaneous amplification and detection of target DNA using amplification primers and a fluorescently labelled detector probe. The presence or absence of applicable and specific DNA is determined by calculating the peak fluorescence (maximum relative fluorescence units) over the course of the amplification process and comparing this measurement to a predetermined threshold value. An extraction control is also used throughout the process.

In addition to the *N. gonorrhoeae* Q^X and *C. trachomatis* Q^X assays, BD also offers the Onclarity **HPV** assay for use on the BD Viper LT system and the BD COR PX/GX System. On the Viper LT system, the HPV assay uses SDA technology for the qualitative detection of 14 high-risk HPV types in a single analysis. Cervical specimens collected by a clinician using an endocervical brush and spatula combination or broom placed in a BD SurePath vial are required. The test specifically identifies types 16, 18 and 45 while concurrently detecting the other high-risk HPV types that include 31, 33, 35, 39, 51, 52, 56, 58, 59, 66 and 68. The assay is indicated for use in, among others, *(1)* women aged 21 years and older with ASC-US cervical cytology test results to determine the need for referral to colposcopy, *(2)* women aged 21 years and older with ASC-US cervical cytology tests results to detect high-risk genotypes 16, 18 and 45, *(3)* women aged 30 years and older to adjunctively screen to detect high-risk HPV types and *(4)* women aged 30 years and older to detect the high-risk HPV genotypes 16, 18 and 45. Like the Q^X *N. gonorrhoeae* and Q^X *C. trachomatis* assays, the BD Onclarity HPV assay is based on the simultaneous amplification and detection of target DNA using amplification primers and a fluorescently labelled detector probe.

In addition to the *C. trachomatis* Q^x and **N. gonorrhoeae** Q^x Amplified DNA assays, BD also offers two additional assays for use on its BD Viper System with XTR technology: the BD ProbeTec **HSV-1** and **HSV-2** Q^x Amplified DNA assay and the BD ProbeTec TV Q^x Amplified DNA assay. The HSV-1 and HSV-2 Q^x assay is a qualitative in vitro diagnostic test that uses SDA technology for the detection and differentiation of HSV-1 and HSV-2 DNA extracted from anogenital lesion specimens. Amplification and detection of target DNA are carried out simultaneously using separate microwells for HSV-1- and HSV-2-specific reactions. The presence or absence of HSV DNA is then determined by calculating the maximum relative fluorescence units over the course of the amplification process and comparing them to a predetermined threshold value. The ***T. vaginalis*** Q^x assay is for the direct and qualitative detection of *T. vaginalis* DNA in clinician-collected female endocervical swab specimens, patient-collected vaginal swab specimens (in a clinical setting), and female urine specimens. The assay is indicated for use with asymptomatic and symptomatic females to aid in the diagnosis of trichomoniasis. The *T. vaginalis* Q^x assay uses SDA technology in a similar way to the assays described above.

BD Viper LT System: The **N. gonorrhoeae** Q^X, ***C. trachomatis*** Q^X and Onclarity **HPV** assays can be performed on the BD Viper LT system, which provides fully automated and integrated molecular testing on a tabletop analyser. The instrument uses homogeneous SDA as the amplification method to test for the presence of pathogens by their genetic content in clinical specimens. The system uses four fluorescence channels with advanced optics and LED light sources for detection, and a thermocycler with 24 control zones designed to ensure uniform temperature control. With less than 15 minutes of hands-on time, the Viper LT system provides up to 30 sample results in 3.5 hours and 120 results per day. As such, the system is designed for a low-to-medium-throughput laboratory.

BD Viper System with XTR technology: The **N. gonorrhoeae** Q^X, ***C. trachomatis*** Q^X, **HSV-1** and **HSV-2** Q^X and ***T. vaginalis*** Q^X assays can be performed on the BD Viper System with XTR technology, which offers fully automated molecular testing, consisting of SDA with rPCR amplification and detection. The system offers continuous batching, with a batch size of 96 specimens. It is a high-throughput system that can provide up to 184 test results every 2 hours and 40 minutes after the first results. The Viper System also provides a closed solid-barrier amplification system to prevent contamination and solid (disposable tips) and neutralized liquid waste for waste management. The system allows for full walkaway operation.

BD MAX system (BD Diagnostics): BD offers several bacterial detection assays for use on its BD MAX system. One of these is a ***C. trachomatis*/N. gonorrhoeae/*T. vaginalis*** assay. The BD MAX system incorporates automated DNA extraction and PCR for the direct and qualitative detection of DNA from *C. trachomatis*, **N. gonorrhoeae** and *T. vaginalis*. The assay may be used to detect *C. trachomatis* or gonococcal DNA in male urine specimens, and *C. trachomatis*, gonococcal or *T. vaginalis* DNA in female urine specimens, clinician-collected female endocervical swab specimens and patient-collected vaginal swab specimens (in clinical settings). The assay is indicated for use to aid in diagnosing chlamydial urogenital disease, gonococcal urogenital disease or trichomoniasis in asymptomatic and symptomatic individuals.

The BD MAX platform automates sample preparation, including target lysis, DNA extraction and concentration, reagent rehydration and target nucleic acid amplification using real-time PCR. The amplified DNA targets are detected using hydrolysis (TaqMan) probes, labelled at one end with a fluorescent reporter dye (fluorophore) and at the other end with a quencher moiety. The system software automatically interprets the test results. The BD MAX platform is capable of batch processing and analysing up to 24 specimens simultaneously. Test results generally take about 2.5–3 hours with an additional 15–20 minutes of hands-on time for completing 24 specimens.

BD COR PX/GX System: The PX instrument automates specimen preparation for testing. It accepts sealed vials and collection devices, including BD SurePath and Hologic PreservCyt LBC primary vials, COPAN FecalSwab and BD collection devices for IVD assays, extracts the required amount of sample and re-seals the container before prewarming and cooling the sample as needed, then delivering it to the GX or MX analytical modules. It taxes approximately 15 minutes to load the module for testing. The GX instrument automatically performs the extraction, amplification and detection required for the BD Onclarity HPV assay, including extended genotype testing. The GX provides 6–8 hours of walkaway system processing time; up to 180 sample results on the BD COR GX module can be performed within one 8-hour shift. The MX instrument automates testing for a range of essential assays for women's health and gastrointestinal testing using a multiplex PCR assay design. The MX can perform up to nine different assays and allows 1700 specimens to be loaded with on-board capacity for reagents and samples that provide more than 6 hours walkaway time and up to 1000 sample results in 24 hours. The first available test for use on the MX instrument is the BD CTGCTV2, which detects *C. trachomatis*, *N. gonorrhoeae* and *T. vaginalis* in a single test. Additional assays for the MX module, including a vaginal panel, are currently in development.

ExiStation Molecular Diagnostic System (Bioneer Corporation): Bioneer offers multiple in vitro diagnostic assays for use on its molecular diagnostic system, ExiStation. The system uses real-time PCR. The following assays for STIs are included:

- AccuPower ***C. trachomatis*** and ***N. gonorrhoeae*** Real-Time PCR Kit for the simultaneous detection of *C. trachomatis* and *N. gonorrhoeae* DNA in human specimens such as urine and vaginal swabs;
- AccuPower ***C. trachomatis*** Real-Time PCR Kit for the detection of *C. trachomatis* DNA in human specimens such as urine, vaginal and urethral swabs;
- AccuPower STI8A-Plex Real-Time PCR Kit for the simultaneous detection of ***C. trachomatis***, ***N. gonorrhoeae***, *Ureaplasma urealyticum* and ***M. genitalium*** DNA in human specimens such as urine and vaginal swabs;
- AccuPower STI 8B-Plex Real-Time PCR Kit for the simultaneous detection of ***T. vaginalis***, *Mycoplasma hominis*, **HSV-1** and **HSV-2** DNA in human specimens such as urine and vaginal swabs;
- AccuPower STI4C-Plex Real-Time PCR Kit for the simultaneous detection of Candida albicans, Gardnerella vaginalis, Ureaplasma parvum and ***T. vaginalis*** DNA in human specimens such as urine and vaginal swabs;
- AccuPower **HPV** 16 & 18 Real-Time PCR Kit for the simultaneous detection of HPV types 16 and 18 DNA in human cervical swab samples;

- AccuPower hrHPV Genotyping & Screening kit for the simultaneous detection of the DNA of 14 **high-risk HPV** types (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68) from human cervical swab and liquid-based cytology specimens.

With the exception of the AccuPower *C. trachomatis* Real-Time PCR Kit and the *C. trachomatis*/*N. gonorrhoeae* combo kit, these assays are CE-IVD-marked. They are designed to be performed on the ExiStation Universal Molecular Diagnostic System, which consists of a nucleic acid extractor, ExiPrep 16Dx, and a real-time PCR analyser, Exicycler 96. The system has flexible configurations, for example, combining two ExiStation instruments with one Exicycler analyser for higher-throughput needs.

In addition, Bioneer offers its ExiStation 48, which consists of the ExiPrep 48Dx and the Exicycler 96 for laboratories with lower-throughput needs. It also offers the ExiPrep 96 Lite, a high-throughput nucleic acid extractor that uses magnetic rods. The instrument can extract nucleic acid from a variety of samples, including serum, plasma, urine, sputum, swabs and more.

ELITe MGB InGenius System (ELITechGroup Solutions): ELITechGroup offers a number of IVD kits and panels. Of relevance to this report is the STI ELITe MGB panel, which is a triplex real-time PCR assay designed to detect and differentiate *C. trachomatis*, *N. gonorrhoeae* and *M. genitalium* DNA in urine and cervical swabs (CE-IVD-marked but not yet available). With regard to *M. genitalium*, the assay can be used in combination with the Macrolide-R/MG ELITe MGB kit to detect macrolide resistance. The assays can be performed on the ELITe InGenius system, which is a sample-to-result molecular diagnostics platform.

The ELITe InGenius platform is an integrated benchtop instrument that automatically performs extraction, real-time PCR amplification and results interpretation. The platform has bidirectional connectivity that enables the laboratory to automatically communicate with an laboratory information system to import testing information and export patient results. Overall turnaround time from extraction to result analysis is approximately 2.5 hours. Hands-on time is about 2 minutes per sample. The ELITe InGenius platform offers random access. One to 12 samples can be processed in parallel in independently controlled real-time PCR units. The platform has a minimum six-plex target capability, which is enhanced with melt curve analysis. The user can potentially mix all kinds of sample matrices and use diverse thermal profiles and even different PCR chemistries at the same time.

FluoroType system (Hain Lifescience, a Bruker Company): Hain Lifescience offers a series of in vitro diagnostic tests, such as its FluoroType assays, which can be performed on its FluoroCycler PCR instrument. The STI assays include FluoroType ***N. gonorrhoeae*** (to detect the microorganism from urethral or cervical swabs, as well as urine) and ***C. trachomatis*** (to detect the microorganism from cervical swabs, urine and ejaculate).

The Hain Lifescience system includes a 96-well format for nucleic acid extraction, the GenoXtract 96 instrument and an amplification instrument, the FluoroCycler 96, which performs subsequent amplification and detection using quantitative PCR (qPCR). The company also offers a lower-throughput extraction platform and qPCR cycler, the GenoXtract 12 and FluoroCycler 12, respectively, which can process or amplify up to 12 samples at once. The company uses a new amplification and probe technology, linear-after-the-exponential PCR combined with fluorescence "lights on/lights off" probes that tile side by side on the target region. Linear-after-the-exponential PCR is an optimized form of asymmetric PCR, in which a limiting primer and an excess primer are used for exponential amplification of double-stranded DNA, followed by linear amplification of a single strand. Each single-stranded amplified sample can then be detected in real time. With the lights on/lights off probes, fluorescence is either emitted or suppressed, which is reflected through a characteristic fluorescence pattern in melt curve analysis. This allows larger regions of target DNA to be interrogated compared with the conventional method of using a single probe. Finally, the test-specific Fluoro software evaluates the test results and displays them automatically. Turnaround time varies with the assay.

Panther System (Hologic): Hologic manufactures several assays for STIs. They are: the Aptima Combo 2 assay (for ***C. trachomatis/N. gonorrhoeae***), the Aptima ***M. genitalium*** assay, the Aptima ***T. vaginalis*** assay and the Aptima **HSV-1** and **HSV-2** assays. The tests are intended for use on the Panther system.

Each assay run on the Panther system involves three main steps, all of which take place in a single tube: target capture; target amplification by TMA; and detection of amplicons by nucleic acid hybridization (hybridization protection assay [HPA]). A collected specimen is transferred into an appropriate specimen transport tube containing transport solution that helps release the ribosomal RNA (rRNA) target and protects it from degrading during storage. The target rRNA, if present, is isolated by using a specific capture oligomer and magnetic microparticles in a method called target capture. The target capture process isolates and purifies the rRNA from clinical specimens, which are then ready for amplification. For each assay, the target nucleic acid strands amplified differ; a unique set of primers is used for each target molecule. Detection of the rRNA amplification product sequences (amplicon) is achieved using nucleic acid amplification, which generates multiple copies of the RNA amplicon. Detection of the amplicon is then achieved by HPA. During this step, a single-stranded chemiluminescent DNA probe labelled with an acridinium ester molecule combines with the amplicon to form stable RNA–DNA hybrids. The light emitted from the labelled RNA–DNA hybrids is measured as RLUs in a luminometer. Assay results for each patient sample are determined based on the analyte signal-to-cut-off ratio. An internal control monitors capture, amplification, detection and operator or instrument error.

*Aptima Combo 2 assay (**C. trachomatis/N. gonorrhoeae***): The Aptima Combo 2 is a target amplification nucleic acid probe test that uses target capture for in vitro qualitative detection and differentiation of rRNA from *C. trachomatis* or *N. gonorrhoeae*. The test is intended for use in the diagnosis of chlamydial and gonococcal urogenital diseases. The following specimens from both symptomatic and asymptomatic individuals may be used for the test: clinician-collected endocervical, vaginal and male urethral swab specimens; clinician-collected gynaecological specimens collected in PreservCyt solution; patient-collected vaginal swab specimens; female and male urine specimens; and throat and rectal swabs. The principle of the assay is as described above; the Aptima Combo 2 assay replicates a specific region of the 23S rRNA from *C. trachomatis* and a specific region of the 16S rRNA from *N. gonorrhoeae* via DNA intermediates.

Aptima ***M. genitalium*** assay: The Aptima *M. genitalium* assay is an in vitro NAAT for the qualitative detection of rRNA from *M. genitalium* on the fully automated Panther system. It is intended for use as an aid in the diagnosis of *M. genitalium* urogenital infections in female and male patients suspected as having the infection. The assay may be used with the following specimens: clinician-collected and self-collected vaginal swabs (in a clinical setting); clinician-collected endocervical swabs; female and male urine; clinician-collected male urethral swabs; and self-collected penile meatal swabs (in a clinical setting). The TMA reaction of the Aptima *M. genitalium* assay amplifies a specific region of the small ribosomal subunit from *M. genitalium* via DNA and RNA intermediates.

*Aptima **T. vaginalis** assay:* The Aptima *T. vaginalis* assay is an in vitro qualitative molecular assays for the detection of rRNA from *T. vaginalis* to aid in the diagnosis of trichomoniasis using the Panther system. The assay may be used to test the following specimens from symptomatic or asymptomatic women: clinician-collected endocervical swabs; clinician-collected vaginal swabs; and specimens collected in PreservCyt solution. The TMA reaction of the Aptima *T. vaginalis* assay amplifies a specific region of the small ribosomal subunit from *T. vaginalis* via DNA and RNA intermediates and generates RNA amplicon molecules.

Aptima HSV-1 and HSV-2 assay: The Aptima HSV-1 and HSV-2 assay is an in vitro diagnostic molecular assays that uses real-time TMA for the qualitative detection and differentiation of HSV-1 and HSV-2 messenger RNA (mRNA) in clinician-collected swab specimens from anogenital skin lesions. The assay is intended for use with swab specimens placed in Aptima specimen transport medium or in viral transport medium that is immediately diluted into specimen transport medium. The Aptima HSV-1 and HSV-2 assay is intended for use as an aid in the diagnosis of HSV-1 and HSV-2 infections in symptomatic male and female patients. The Aptima HSV-1 and HSV-2 assay is indicated for use on the Panther system.

*Aptima **HPV** assay:* The Aptima HPV assay is an in vitro molecular assay for the qualitative detection of E6/E7 viral messenger mRNA from 14 high-risk types of HPV associated with cervical cancer and precancerous lesions and is intended for use on the Panther system. The high-risk HPV types detected by the assay include 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66 and 68. However, the assay does not distinguish between the 14 high-risk types of HPV. The Aptima HPV assay is performed with Hologic's ThinPrep liquid cytology specimens. The assay is for use in screening women aged 21 years and older with ASC-US cervical cytology results to determine the need for referral to colposcopy and to use adjunctively with cervical cytology to screen women aged 30 years and older to assess the presence or absence of high-risk HPV types.

*Aptima **HPV** 16 18/45 Genotype Assay:* The Aptima HPV 16 18/45 genotype assay is an in vitro molecular assay for the qualitative detection of the E6/E7 viral mRNA of human HPV types 16, 18 and 45 in cervical specimens from women with Aptima HPV assay-positive results. The assay can differentiate HPV 16 from HPV 18 or HPV 45, but does not differentiate between HPV 18 and HPV 45. Cervical specimens in ThinPrep Pap test vials containing PreservCyt solution and collected with broom-type or cytobrush/spatula collection devices may be tested with the Aptima HPV 16 18/45 genotype assay. The assay is intended for use on the Panther system.

Additional assays that can be run on the Panther system include bacterial vaginosis, Candida vaginitis/*T. vaginalis* as well as quantitative assays for HIV-1 viral load, hepatitis B virus and hepatitis C virus.

The Panther system is a molecular diagnostic platform with random-access testing capability on a fully integrated and automated molecular system. Within Panther, all nucleic acid testing steps, from primary sample tube to results, are fully automated in one system with first reportable results within 3 hours after loading the samples, and five results every 5 minutes thereafter. Samples can be continuously loaded with up to 120 samples at a time. Reagent controls and calibration are valid for 24 hours. At least 275 samples can be run within an 8-hour shift, or 500 in a 12-hour period (an additional 225 samples can be run without operator attendance). Four reagent lanes allow up to four Aptima test kits to be on-board and randomly accessed at any time: this could be four kits of the Combo 2 assay or any combination of the other molecular diagnostic assays available on Panther, including the HIV-1 Quant Dx, *T. vaginalis*, HPV and HPV genotyping, HCV Quant Dx, HBV Quant and HSV-1 and HSV-2 assays. In addition, the Panther system is scalable. With the addition of the Panther Fusion module, PCR testing, as well as TMA and TMA with reverse transcription assays can all be run on a single, fully automated platform.

Larger laboratories can also add the Panther Plus module to increase capacity and walkaway time. Another available module is the Panther Link, which virtually connects the Panther systems and enables sharing of information and monitoring the laboratory from a centralized dashboard.

Alethia Molecular Diagnostic System (Meridian Bioscience): Meridian Bioscience offers the Alethia **HSV-1** and **HSV-2** assay. It is an in vitro diagnostic test for the qualitative detection and differentiation of HSV-1 and HSV-2 DNA in cutaneous and mucocutaneous lesion specimens from male and female patients suspected of herpes infection. The assay is intended for use on its Alethia platform, which, it should be noted, the company indicates is only for use in hospital, reference or state laboratory settings and not for use at POC. The assay uses LAMP technology to detect HSV-1 and HSV-2 by targeting segments of the HSV-1 and HSV-2 genomes. K

These assays are intended to be performed on the Alethia platform, which is a compact, automated isothermal amplification and detection system. There is a separate sample preparation device. The Alethia is a menu-driven laboratory instrument with two independent sample-processing blocks, identified as A and B. Sample heating and optical detection is carried out for up to five two-chambered illumigene devices per block. Each two-chambered Alethia device contains a sample chamber and a control chamber. Amplification of target DNA occurs during the heat cycle and results in the formation of precipitate detected by the Alethia optics system. The precipitate generated by the presence of amplified target DNA leads to a turbid sample or control reaction solution, which is then measured using absorbance. One to 10 qualitative results are available in less than an hour on the Alethia instrument.

QIAsymphony SP/AS and Rotor-Gene Thermocyclers (QIAGEN): QIAGEN manufactures a series of artus assay kits. The kits are designed to be used with the automated extraction and sample preparation system QIAsymphony SP/AS and must then be run on one of the QIAGEN real-time Rotor-Gene Q (RGQ) thermocyclers for amplification and detection.

Of relevance to this report are two assays, the artus ***C. trachomatis/N. gonorrhoeae*** QS-RGQ kit and the artus **HSV-1/HSV-2** RGQ kit. The artus *C. trachomatis*/*N. gonorrhoeae* assay is an in vitro molecular test for the qualitative detection of *C. trachomatis* plasmid and genomic DNA, and *N. gonorrhoeae* genomic DNA from vaginal swabs and urine. The artus HSV-1/HSV-2 assay is also an in vitro molecular test, but is for the qualitative detection of HSV-1 and HSV-2 from human cerebrospinal fluid and plasma. Both kits, which come ready to use with all optimized reagents required to run the tests, use PCR and are configured for use with the QIAsymphony SP/AS and RGQ instruments.

The QIAsymphony SP/AS instruments provide automated sample preparation and assay set-up. The QIAsymphony SP can process 1–96 samples (in batches of 24) with sample volumes up to 1 ml. It is a ready-to-run instrument that requires minimal installation. The SP can be combined with the QIAsymphony AS device in a fully integrated system that can automate the entire workflow. To reduce manual handling and minimize the risk of sample contamination, samples processed on the SP can be transferred automatically to the AS, or the two instruments can be operated independently. Despite their ease of use, the QIAsymphony system and RGQ instruments are designed for use in sophisticated laboratories.

The QIAGEN real-time PCR cycler, the RGQ, offers a unique centrifugal rotary design. Each tube in the instrument spins in a chamber of moving air, which keeps all samples at precisely the same temperature during rapid thermal cycling. Detection is also uniform. When each tube aligns with the detection optics in the instrument, the sample is illuminated and the fluorescence signal is rapidly collected from a single short optical pathway. As stated by the manufacturer, the thermal and optical uniformity of the system results in sensitive, precise and fast real-time PCR.

digene Hybrid Capture 2 (HC2) Technology and Rapid Capture System (RCS) (QIAGEN): QIAGEN offers several STI assays using the digene HC2 technology, which uses nucleic acid hybridization with signal amplification and microplate chemiluminescence for the qualitative detection of pathogens. One of these assays is the digene HC2 HPV DNA test, which is an in vitro diagnostic test for the qualitative detection of 18 low-risk and high-risk types of HPV DNA in cervical specimens, including biopsies. When using the low-risk and high-risk HPV probes, the test is indicated: *(1)* to aid in the diagnosis of sexually transmitted HPV infections with HPV types 6, 11, 16, 18, 31, 33, 35, 39, 42, 43, 44, 45, 51, 52, 56, 58, 59, and 68; and *(2)* to differentiate between two HPV DNA groups: low-risk HPV types 6, 11, 42, 43, and 44; and high-risk HPV types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, and 68. It should be noted, however, that the specific HPV type present cannot be determined.

In addition to the *digene* HC2 DNA test, QIAGEN offers several assays for the detection of ***C. trachomatis*** and ***N. gonorrhoeae*** infections. These are the *digene* HC2 *C. trachomatis*/*N. gonorrhoeae* test, HC2 *C. trachomatis*-ID DNA test, HC2 *N. gonorrhoeae*-ID DNA and HC2 *C. trachomatis-N. gonorrhoeae* Dual ID DNA test. All of the assays are in vitro nucleic microplate assays based on signal-amplified nucleic acid hybridization that uses chemiluminescence to detect specific DNA. The *digene* HC2 *C. trachomatis*/*N. gonorrhoeae* test is indicated for the combined qualitative detection of DNA from *C. trachomatis* and *N. gonorrhoeae* from cervical specimens and is intended as an initial test to identify symptomatic or asymptomatic women with *C. trachomatis* or gonococcal infection. Follow-up testing with the HC2 *C. trachomatis*-ID DNA test and the HC2 *N. gonorrhoeae*-ID DNA tests is required to identify the microorganism(s)

present in a specimen that tests positive with the combined test. The HC2 *C. trachomatis*-GC Dual ID DNA test is for the separate detection of *C. trachomatis* and gonococcal infections in the same sample and is designed for use as a screening assay for *C. trachomatis* and *N. gonorrhoeae* in patient populations.

All of the above *digene* DNA tests operate on the same principle. Specimens containing the target DNA hybridize with an assay-specific RNA probe or probes. The resultant RNA–DNA hybrids are captured onto the surface of a microplate well coated with antibodies specific for RNA–DNA hybrids. Immobilized hybrids are then reacted with ALP-conjugated antibodies specific for RNA–DNA hybrids, and detected with a chemiluminescent substrate. Several ALP molecules are conjugated to each antibody. Multiple conjugated antibodies bind to each captured hybrid resulting in substantial signal amplification. As the substrate is cleaved by the bound ALP, light is emitted, which is measured as RLUs on a luminometer. The intensity of the light emitted denotes the presence or absence of target DNA in the specimen.

High-volume sample-throughput testing with each of the *digene* DNA assays described can be performed using the QIAGEN RCS. The system is a robotic microplate processor consisting of microprocessor-controlled components. The system is controlled using operating software that resides on the hard drive of a required personal computer interfaced with the RCS. The RCS can handle up to 352 specimens in an 8-hour shift, including a 3.5-hour walkaway period. Up to 704 specimen results can be generated sequentially in 13 hours. Operator intervention is required for specimen preparation, loading of specimen racks onto the deck, deck set-up, chemiluminescence signal detection, and result reporting. The RCS is U.S. FDA-approved for use with the digene HC2 High-Risk HPV DNA, digene HC2 *C. trachomatis*/*N. gonorrhoeae* DNA, digene HC2 *C. trachomatis*-ID DNA and digene HC2 *N. gonorrhoeae*-ID DNA tests.

NeuMoDx Molecular System (QIAGEN N.V.): NeuMoDx Molecular, now a subsidiary of QIAGEN, offers a continuous, random-access real-time PCR system with a wide range of assays. The system consists of a family of scalable platforms, including the NeuMoDx 288 and NeuMoDx 96 instruments that fully connect the entire molecular diagnostic process from sample to result. Depending on the assay, turnaround time is as little as 40 minutes.

NeuMoDx offers assays for ***C. trachomatis/N. gonorrhoeae***, ***T. vaginalis/M. genitalium*** and **HPV**. The *C. trachomatis*/*N. gonorrhoeae* assay uses urine as its specimen and the system performs all the steps required to extract the target nucleic acid, prepare the isolated DNA for real-time PCR amplification, and if present, amplify and detect the products of amplification (sections of the targeted gene sequences of the *C. trachomatis* and *N. gonorrhoeae* chromosomes and plasmids). Similarly, the NeuMoDx *T. vaginalis*/*M. genitalium* assay is designed to detect and differentiate *T. vaginalis* and *M. genitalium* DNA simultaneously. The assay targets the region encoding a hypothetical protein (TVAG_305840) in the *T. vaginalis* genome and the sequences encoding the IgG-blocking protein M and thymidylate kinase in the *M. genitalium* genome. A urine specimen is required. Finally, the NeuMoDx HPV is a fully automated, in vitro, real-time PCR-based nucleic acid amplification assay for the qualitative detection of high-risk types of HPV DNA in cervical specimens. The test specifically identifies HPV-16 and HPV-18 while concurrently detecting the other high-risk types (31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 67 and 68) at clinically relevant infection levels. Cervical specimens that may be tested with the NeuMoDx HPV Assay include cervical specimens collected using a brush/broom-type collection device (collected by a physician).

cobas 4800 system (Roche): Roche manufactures several qualitative molecular tests for sexual health for use on its cobas 4800 system. These include the cobas ***C. trachomatis/N. gonorrhoeae*** v2.0 assay, the cobas 4800 **HPV** assay and the cobas **HSV-1/HSV-2** assay.

cobas ***C. trachomatis/N. gonorrhoeae*** v2.0 Assay: The cobas *C. trachomatis*/*N. gonorrhoeae* v2.0 assay is an automated, qualitative, IVD test that uses real-time PCR and nucleic acid hybridization for the qualitative detection of *C. trachomatis* and *N. gonorrhoeae* DNA from vaginal swab specimens self-collected in a clinical setting and male urine from both symptomatic and asymptomatic individuals. Specimens are collected in cobas PCR media. The cobas *C. trachomatis*/*N. gonorrhoeae* v2.0 assay detects DR-9, a direct repeat region and target of the *N. gonorrhoeae* assay. It also simultaneously detects two *C. trachomatis* independent DNA targets, one in the cryptic plasmid and the other on the *C. trachomatis* genome. This design can detect infections caused by wild-type *C. trachomatis*, the Swedish variant (nvCT) and other Chlamydia strains that may harbour deletions in the cryptic plasmid, or those that do not carry the cryptic plasmid.

*cobas **HPV** assay:* The cobas HPV assay is a qualitative in vitro test for the detection of HPV in cervical specimens collected by a clinician using an endocervical brush/spatula and placed in the ThinPrep Pap test PreservCyt solution or using a cervical broom and placed in SurePath preservative. The assay is indicated for use in routine cervical cancer screening according to the applicable professional medical guidelines and HPV primary screening of women to assess the risk for cervical precancer and cancer. The test uses amplification of target DNA by PCR and nucleic acid hybridization to detect 14 high-risk HPV types, including genotypes 16 and 18, the two genotypes responsible for about 70% of all cervical cancers, in a single analysis.

*cobas **HSV-1** and **HSV-2** assay:* The cobas HSV-1 and HSV-2 assay is an automated, qualitative IVD using real-time PCR for the direct detection and differentiation of HSV-1 and HSV-2 from DNA in clinician-collected, external anogenital lesion specimens from symptomatic male and female patients. Specimens are collected in the MSwab Collection, Transport and Preservation System

from symptomatic patients. The cobas HSV-1 and HSV-2 consists of two primary processes: *(1)* automated sample preparation to extract nucleic acids from specimens; and *(2)* PCR amplification of target DNA sequences using HSV-1- and HSV-2-specific primers, and real-time detection cleaved fluorescently labelled HSV-1- and HSV-2-specific oligonucleotide detection probes. An internal control containing an unrelated randomized DNA sequence is added to all samples before automated sample preparation and is amplified and detected simultaneously with each sample to monitor the process.

The cobas 4800 system fully integrates automated total nucleic acid isolation directly from primary and secondary tubes, automated PCR set-up and real-time PCR. It is intended for laboratories with a medium workflow. The system consists of the cobas x480 instrument and the cobas z480 analyser. According to Roche, it features minimal hands-on time (30 minutes for a run of 24–96 samples). It can take multiple sample types, can detect multiple test targets and has what the manufacturer describes as an intuitive workflow.

cobas 6800/cobas 8800 systems (Roche Molecular Diagnostics): Roche offers a number of commercially available assays related to sexual health for use on its cobas 6800/8800 systems. These include the cobas ***C. trachomatis/N. gonorrhoeae*** assay, the cobas ***T. vaginalis/M. genitalium*** assay and the cobas **HPV** assay.

*cobas **C. trachomatis/N. gonorrhoeae** assay:* The cobas *C. trachomatis*/*N. gonorrhoeae* assay is an automated, qualitative in vitro molecular assay that uses real-time PCR for direct detection of *C. trachomatis* or *N. gonorrhoeae* DNA in male and female urine, clinician-instructed self-collected vaginal swab specimens (collected in a clinical setting), clinician-collected vaginal swab specimens and endocervical swab specimens, all collected in cobas PCR medium, and cervical specimens collected in PreservCyt solution. The test is intended as an aid in diagnosing chlamydial and gonococcal disease in both symptomatic and asymptomatic individuals. Target-specific primers and two probes are used to detect, but not discriminate between, the *C. trachomatis* cryptic plasmid and the ompA gene.

Additionally, target-specific primers and two probes are used to detect, but not discriminate between, two conserved sequences in the *N. gonorrhoeae* DR-9 region. A DNA internal control, used to monitor the entire sample preparation and PCR amplification process, is introduced into each specimen during sample processing.

*cobas **T. vaginalis/M. genitalium*** assay: The cobas *T. vaginalis*/*M. genitalium* assay is an automated, qualitative, in vitro molecular assay that uses PCR for the direct detection of *T. vaginalis* and *M. genitalium* DNA in male or female urine, self-collected vaginal swab specimens (collected in a clinical setting), clinician-collected vaginal swab specimens and endocervical specimens, all collected in cobas PCR medium. The assay can also detect *T. vaginalis* DNA in cervical specimens (collected in a PreservCyt solution) and *M. genitalium* DNA in self-collected meatal swab specimens (collected in a clinical setting), and clinician-collected meatal swab specimens. The cobas *T. vaginalis*/*M. genitalium* assay is intended as an aid in the diagnosis of *T. vaginalis* and *M. genitalium* infections in individuals suspected to have either infection. A vaginal swab (either self-collected or clinician-collected) is the preferred specimen type for *M. genitalium* testing in females because of its higher sensitivity compared to endocervical swabs and urine. For males, urine is the preferred specimen type because of higher sensitivity compared to meatal swabs. The cobas *T. vaginalis*/*M. genitalium* assay is based on fully automated sample preparation followed by PCR amplification and detection. Selective amplification of target nucleic acid from the sample is achieved by the use of target-specific forward and reverse primers for *T. vaginalis* and *M. genitalium*, which are selected from highly conserved regions within the respective target microorganism. A thermostable DNA polymerase enzyme is used for PCR amplification. Real-time detection and discrimination of PCR products is achieved by measuring the fluorescence of the released reporter dyes for the *T. vaginalis* and *M. genitalium* targets and DNA internal control, respectively.

*cobas **HPV** assay:* The assay has already been described in the cobas 4800 system. The assays for use on the cobas 6800/8800 systems are U.S. FDA-cleared and CE-IVD-marked. The systems offer the fastest time to results with the highest throughput available. The cobas 6800/8800 systems are fully automated solutions designed for donor screening, viral load monitoring, as well as women's health and microbiology testing.

The 6800/8800 systems are available in medium- and high-throughput models. Each system provides results for the first 96 tests in less than 3.5 hours, with the 6800 system delivering up to 384 results in an 8-hour shift, and the 8800 system generating up to 960 results in the same amount of time. Both systems also allow for simultaneous processing of multiple assays and are designed to enable up to 8 hours (cobas 6800) and 4 hours (cobas 8800) of walkaway time with minimal user interaction.

Allplex, Anyplex and Seeplex systems (Seegene): Seegene offers several highly multiplexed molecular assay kits that use real-time PCR or capillary electrophoresis for amplicon detection. These include the Allplex, Anyplex and Seeplex test kits, many of which are CE-IVD-marked. The company does not supply completely integrated sample-to-result systems. For example, sample preparation is not provided by Seegene; therefore, a product from another manufacturer must be used for this purpose. While DNA extraction and an initial PCR amplification step can be run on Seegene instruments (SEEPREP12 and SEEAMP, respectively), additional real-time PCR amplification steps or automated electrophoresis, depending on the assay kit, must be done on other systems validated for use with Seegene kits. For real-time PCR, these include the ABI 7500 Real-Time PCR (Thermo Fisher Scientific) and the CFX96 Real-Time PCR (Bio-Rad Laboratories).

Seegene has developed proprietary software, Multiple Detection Temperatures Technology, to discriminate between the 10 channels on the CFX96 platform. It allows simultaneous identification and quantification of multiple pathogen targets in a single channel without melt curve analysis after amplification. In addition, the viewer software analyses the raw data to generate test results from the various Seegene assays. The multiplicity of steps and equipment required to perform the Seegene test kits suggests that they should be used only in the most sophisticated laboratory settings:

- The Seegene Allplex assays include two STI assays: the STI Essential assay, which tests for ***C. trachomatis***, ***N. gonorrhoeae***, ***T. vaginalis***, ***M. genitalium***, *M. hominis*, *U. parvum* and *U. urealyticum*, and an assay that tests for ***C. trachomatis***, ***N. gonorrhoeae***, ***T. vaginalis*** and ***M. genitalium***;
- The Seegene Anyplex assays include three STI assays, the Anyplex II STI-5, which detects ***T. vaginalis***, ***M. genitalium***, *M. hominis*, *U. parvum* and *U. urealyticum*; the Anyplex II STI-7, which detects ***C. trachomatis***, ***N. gonorrhoeae***, ***T. vaginalis***, ***M. genitalium***, *M. hominis*, *U. parvum* and *U. urealyticum*; and the Anyplex ***C. trachomatis/N. gonorrhoeae*** real-time assay;
- The Seegene Seeplex assays include two STI assays, the Seeplex STD6 ACE Detection, which tests for ***C. trachomatis***, ***T. vaginalis*** and *M. hominis*, and the Seeplex STI Master Panel 1, which detects ***C. trachomatis***, ***T. vaginalis*** and *M. hominis*;
- Other CE-IVD-marked assays include multiplex respiratory pathogen assays, **HSV-1** and **HSV-2**, **HPV** (screening and genotyping) and meningitis.

Other open systems

AmpliSens (InterLabService): InterLabService provides a variety of PCR kits for the detection of infectious diseases. These include AmpliSens kits for ***C. trachomatis***, ***N. gonorrhoeae***, ***T. vaginalis***, ***M. genitalium***, *U. urealyticum*, *U. parvum*, G. vaginalis, **HSV-1** and **HSV-2**. The company also manufactures kits for both high-risk and low-risk **HPV**. The assays must be performed on instruments for which they have been adapted, which, depending on the assay, may include the Rotor-Gene 3000 or Rotor-Gene 6000 (Corbett Research) and the iCycler or iCycler 5 (Bio-Rad Laboratories), among others.

EasyScreen (Genetic Signatures): Genetic Signatures offers a wide array of assays based on real-time PCR for the qualitative detection of various sexually transmitted pathogens, including ***C. trachomatis***, ***N. gonorrhoeae***, ***T. vaginalis***, ***M. genitalium***, *U. urealyticum*, *U. parvum*, **HSV-1** and **HSV-2**, among others. Assays must be performed on instruments with which they are compatible; these include a variety of laboratory-based real-time PCR instruments.

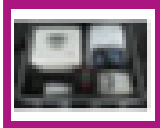
Table 23: Molecular multiplex platforms for use at POC or near-POC for the detection of sexually transmitted infections

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
AmplexDiagnostics eazyplex Complete STDs (*C. trachomatis*, *N. gonorrhoeae*, *M. genitalium*), *C. trachomatis*, *C. trachomatis*/*N. gonorrhoeae* (Genie III platform) 	Qualitative	N/A	Urethral, rectal/anal, vaginal, cervical, and pharyngeal swab	CE-IVD	N/A
binx io *C. trachomatis*/*N. gonorrhoeae* assay (io system)  	Qualitative	30 minutes	Vaginal swab, male urine	FDA CE-IVD CLIA-waived	N/A

Table 23 (continued): Molecular multiplex platforms for use at POC or near-POC for the detection of sexually transmitted infections

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
Bosch Healthcare Solutions STI Multiplex Array/ Vivalytic Analyzer (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HSV) 	Qualitative	2 hours 30 minutes	Urogenital swab, urine specimen	CE-IVD	Biochip array
Cepheid Xpert *C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis* assays (GeneXpert System) 	Qualitative	<1 hour up to 90 minutes	Urine, rectal swab, throat swab, vaginal swab (*C. trachomatis*/*N. gonorrhoeae*); endocervical swab (*C. trachomatis*/*N. gonorrhoeae*, HPV); male and female specimen (*T. vaginalis*)	FDA (*C. trachomatis*/*N. gonorrhoeae*, *T. vaginalis*) CE-IVD (*C. trachomatis*/*N. gonorrhoeae*, HPV, *T. vaginalis*)	N/A
HiberGene Diagnostics HG *C. trachomatis*/*N. gonorrhoeae* Combo test, HSV test, *M. genitalium* test (HG Swift and HG Swift Plus) 	Qualitative	< 40–60 minutes for positive, 50–70 minutes for negative	Vaginal swab, urine (*C. trachomatis*/*N. gonorrhoeae*, *M. genitalium*), Urogenital swab (HSV)	CE-IVD	LAMP
Luminex ARIES HSV-1 and HSV-2 assay (ARIES and ARIES M1 Systems) 	Qualitative	2 hours	Cutaneous or mucocutaneous lesion	FDA CE-IVD	Real-time PCR

Table 23 (continued): Molecular multiplex platforms for use at POC or near-POC for the detection of sexually transmitted infections

Manufacturer/test (device)	Reporting	Turnaround time	Sample type	Approval status	Test technology
Molbio Diagnostics Truenat assays (*C. trachomatis*, *C. trachomatis*/*N. gonorrhoeae*, HPV, *N. gonorrhoeae*, TV) **(Truelab Real Time microPCR System)**  	Semi-quantitative	35 minutes to 1 hour (HPV)	Endocervical swab, vaginal swab, male urethral swab, urine (*C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*); cervical specimen (HPV)	CE-IVD	microPCR
QuidelOrtho SOLANA HSV-1 + HSV-2/varicella zoster virus and Trichomonas Assays 	N/A	50 minutes	Vaginal swab, urine	N/A	HDA
QuidelOrtho Lyra Direct HSV-1 + HSV-2/varicella zoster virus assay	N/A	Less than 70 minutes	Cutaneous or mucocutaneous swab	N/A	PCR
QuidelOrtho AmpliVue HSV-1 + HSV-2 assay 	Qualitative	60 minutes	Cutaneous or mucocutaneous swab	N/A	Lateral flow strip
Ustar Biotechnologies *C. trachomatis*/*N. gonorrhoeae*, *M. genitalium* and *T. vaginalis* DNA CPA assay; EasyNAT HPV 6/11, HSV-1 and HSV-2 assays **(EasyNAT Platform)**  	Qualitative	50 minutes	Male urinary tract swab, cervical swab (*C. trachomatis*, *N. gonorrhoeae*); endocervical swab (HPV); urogenital swab (HPV, HSV, *T. vaginalis*); urine (*M. genitalium*, *T. vaginalis*)	CE-IVD N/A (HPV)	CPA
Visby Medical C. trachomatis/N. gonorrhoeae/T. vaginalis test 	Qualitative	50 minutes	Vaginal swab	FDA CLIA-waived	RT–PCR

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CLIA: Clinical Laboratory Improvement Amendments waiver; CPA: cross priming amplification; FDA: U.S. Food and Drug Administration; HDA: helicase-dependent amplification; LAMP: loop-mediated isothermal amplification; N/A: not applicable; PCR: polymerase chain reaction; RT–PCR: PCR with reverse transcription.

Molecular multiplex platforms for use at POC or near-POC for detection of STIs

eazyplex (AmplexDiagnostics): AmplexDiagnostics offers eazyplex lyophilized ready-to-use amplification test kits validated for use on the Genie II platform (OptiGene). The eazyplex tests are qualitative in vitro molecular diagnostic tests to detect bacterial DNA in no more than 30 minutes. No extraction is required and kits can be stored at ambient temperature.

The test process is as follows. Samples are suspended in resuspension and lysis fluid buffer solution and incubated for 2 minutes with thermal lysis. Then, 25 ml of the resuspension and lysis fluid is added to each tube of the strip containing ready-to-use mastermix. The test strip is then immediately placed into the Genie II instrument, where it is incubated at 66 °C for 30 minutes with fluorescence monitoring. Isothermal amplification is indicated by a strong increase in fluorescence signal in the form of a typical amplification curve. Different colours are given to each of the tested gene variants. Genie II has two heating blocks, each of which can process a single eight-microtube test strip. The blocks can be controlled independently or run together to process up to 16 samples.

It is a fully portable, compact and lightweight platform for target isothermal amplification and detection designed for use at or near-POC and suitable for use in demanding environments. The platform includes dual-channel fluorescence measurement to allow the use of internal controls and multiplexed assays. It also has positional information through GPS and offers wireless connectivity in the form of Bluetooth and Wi-Fi. Genie III incorporates a rechargeable lithium polymer battery that can support operation of the instrument for an 8-hour day. The instrument has a single heating block, which can process a single eight-microtube strip:

- eazyplex Complete STDs: This is a multiplex IVD assay for the qualitative detection of ***C. trachomatis, N. gonorrhoeae***, *U. urealyticum*, *M. hominis*, *M. genitalium* and *T. pallidum* from human swabs (urethral; rectal/anal; vaginal; cervical, pharyngeal);
- eazyplex STD ***C. trachomatis/N. gonorrhoeae***: This is an IVD assay for the qualitative detection of *C. trachomatis* and *N. gonorrhoeae* from human swabs (urethral; rectal/anal; vaginal; cervical, pharyngeal);
- eazyplex STD ***C. trachomatis***: This is an IVD assay for the qualitative detection of *C. trachomatis* from human swabs (urethral; rectal/anal; vaginal; cervical, pharyngeal).

binx io system (binx health, formerly Atlas Genetics): The binx *io* platform is a rapid, multiplex, molecular diagnostic system that can deliver laboratory quality results in about 30 minutes. The system, which consists of a small instrument and disposable cartridge, that contains all reagents necessary to run a test, is designed to be easy to use, and is fully automated. The operation of the instrument is designed to be simple and intuitive; the user interacts with the instrument through a touchscreen interface that then guides the user through the io system test process. Once the raw sample has been added to the cartridge and loaded into the instrument, no further interaction is required. The instrument fits easily on a benchtop and is fully integrated, enabling the movement of a sample and reagents within the cartridge. The cartridge has three main assay steps: *(1)* sample preparation to isolate and purify target DNA; *(2)* ultra-rapid PCR, which amplifies specific regions of DNA from the target organisms; and *(3)* a proprietary electrochemical detection to identify the presence of amplified DNA. Once the test is completed, a qualitative "detected/not detected" result is available with no clinical or laboratory interpretation needed.

binx health's core focus on STIs leads with their first application to diagnose two of the most tested for STIs globally: ***C. trachomatis*** and ***N. gonorrhoeae***. The binx health io *C. trachomatis*/*N. gonorrhoeae* assay is intended for use with female vaginal swab specimens, collected either by a clinician or self-collected by a patient in a clinical setting, or male urine specimens, as an aid in the diagnosis of symptomatic or asymptomatic *C. trachomatis* and *N. gonorrhoeae*. The binx test is designed to provide a result directly from an unpurified patient sample in about 30 minutes with equivalent accuracy and performance as current standard-of-care platforms run in central laboratories (which can take seven or more days). The goal is to help provide earlier diagnosis and accurate detection and ultimately more timely treatment to aid in the prevention of onward transmission and the serious consequences that go with undiagnosed and prolonged infection.

GeneXpert System (Cepheid): The Cepheid GeneXpert System is a fully automated and integrated system for PCR-based molecular testing; including a combined *C. trachomatis*/*N. gonorrhoeae* assay for simultaneous detection (Xpert *C. trachomatis*/*N. gonorrhoeae*) and a test for ***T. vaginalis*** (Xpert *T. vaginalis*), which is also U.S. FDA-cleared for symptomatic and asymptomatic men. The company also offers a test for **HPV**, the Xpert HPV.

The Xpert ***C. trachomatis/N. gonorrhoeae*** assay, performed on the GeneXpert System is a qualitative, in vitro, real-time PCR test for automated detection and differentiation of genomic DNA from *C. trachomatis* or *N. gonorrhoeae*. It is CE-IVD-marked and U.S. FDA-cleared. The assay may be used on the following specimens from both asymptomatic and symptomatic patients: female and male urine; endocervical swab; rectal swab; throat swab; and patient-collected vaginal swab (collected in a clinical setting). The test process is straightforward, with total hands-on time estimated to be less than 1 minute. The operator *(1)* obtains either urine or swab samples, which are previously collected and stored in the Cepheid Transport Reagent; *(2)* transfers the sample to the Xpert cartridge; and *(3)* inserts the cartridge into the Xpert system and starts the assay. Time to result is approximately 90 minutes. The performance of the Xpert *C. trachomatis*/*N. gonorrhoeae* assay has been evaluated and found to be very good relative to established laboratory-based assays *(48–50)*.

The Xpert ***T. vaginalis*** molecular assay detects both male and female specimens. The time to result of the test is approximately 1 hour *(51)*.

The Xpert **HPV** assay is a less than 60-minute test for cervical cancer-related human papillomaviruses. It is a multiplexed test that targets the E6 and E7 oncogenes of 14 high-risk HPV types and independently calls out genotype 16, the most common type associated with ICC worldwide *(52)*; a combined call out for genotypes 18 and 45 is also closely associated with ICC *(53)*; 11 other high-risk genotypes are detected in combined channels. Xpert HPV uses samples from endocervical cells collected in PreservCyt solution (Hologic) with either a broom-like device or an endocervical brush and spatula combination.

All of Cepheid's STI tests have a unique sample adequacy control (SAC). Each self-contained cartridge includes a SAC, which detects the presence of a single-copy human gene and monitors whether the sample contains human DNA for enhanced results integrity *(54)*.

The GeneXpert System integrates and automates sample preparation, amplification and detection in a single-use, self-contained cartridge. Most liquids and dry reagents along with enzymes are prefilled so that preanalytical steps are minimized, greatly reducing opportunities for sample mix-ups and operational errors. GeneXpert cartridges can handle a variety of sample volumes (microlitre to millilitre volume range) within macrofluidic chambers and then concentrate the target material down to microfluidic volumes, which can increase the sensitivity of the assays, if needed.

Furthermore, the GeneXpert System is modular. Individual modules contain solid-state circuitry that controls temperature, pressure, rotation of the valve that moves the liquid between reservoirs in the cartridge and the detection software. These individual modules are packaged in cabinets that can hold up to 1, 2, 4, 16, 48 or 80 modules. The latter two systems (Infinity-48 and Infinity-80) are fully automated, walkaway robotic systems developed for high-throughput laboratory applications. Additionally, the modules can be removed and replaced individually so that the entire system is not compromised if one module fails.

The GeneXpert System is sufficiently simple that training can usually be completed within half a day or less. Furthermore, although the system was designed to use AC power, its low wattage requirements allow it to be powered by a 12VDC/120VAC voltage converter in mobile laboratories. It has also been installed in remote clinic sites powered by solar panels. The GeneXpert software comes preinstalled on a desktop or laptop computer and results can be displayed for each module in real time or uploaded via an Internet connection to a central database or institutional logistics management information system. Cepheid C360 allows for system monitoring to observe instrument performance data, and disease surveillance to aggregate and monitor disease state testing data.

HG Swift (HiberGene Diagnostics): HiberGene Diagnostics manufactures the HG Swift and HG Swift Plus instruments for the diagnosis of infectious disease. They are lightweight and compact. Both instruments use isothermal LAMP technology, which uses new primer design, an efficient strand-displacing polymerase, along with dual-channel fluorometric detection and unique results-calling software.

The HG Swift instruments are compact and portable. The HG Swift instrument, which has a single thermal block, can simultaneously detect pathogens across eight tubes in two fluorescence channels; the HG Swift Plus instrument, which has two thermal blocks, can simultaneously detect pathogens across 16 tubes in two fluorescent channels. The system features an integrated touchscreen interface and provides real-time display of amplification. The system can be run with AC mains or battery power. Turnaround time is approximately 40 minutes.

HiberGene has introduced the HG ***C. trachomatis/N. gonorrhoeae*** Combo test using isothermal LAMP technology for its system. The *C. trachomatis*/*N. gonorrhoeae* Combo test is an in vitro test for the detection and differentiation of clinically relevant strains of *C. trachomatis* and *N. gonorrhoeae* in vaginal swabs or urine, which must be cleaned, washed, resuspended and lysed before being run on the HG Swift or HG Swift Plus instrument. Turnaround time is less than 60 minutes for a positive result and approximately 70 minutes for a negative result. No peer-reviewed published evaluations of the HG *C. trachomatis*/*N. gonorrhoeae* Combo test were found.

HiberGene has also recently introduced two additional assays using its isothermal LAMP technology. The first is an assay for the detection of ***M. genitalium*** from a single urine (male or female) or vaginal swab sample. The test follows the same protocol as the HG *C. trachomatis*/*N. gonorrhoeae* Combo test. Turnaround time is less than 60 minutes for a positive result and approximately 70 minutes for a negative result. The second is an assay for the detection of **HSV-1** and **HSV-2** from a urogenital swab sample. Turnaround time is less than 40 minutes for a positive result and approximately 50 minutes for a negative result.

ARIES and ARIES M1 systems (Luminex): Luminex offers the ARIES HSV-1 and HSV-2 assay, which is a real-time PCR-based qualitative IVD test for the direct detection and differentiation of HSV-1 and HSV-2 DNA in cutaneous or mucocutaneous lesion specimens from symptomatic patients.

The ARIES **HSV-1** and **HSV-2** assay is designed for use on the real-time PCR Luminex ARIES system, a two-module instrument, or ARIES M1 system, a single-module instrument, each of which consists of the associated ARIES software, a stool resuspension kit, an assay-specific test cassette and an assay-specific protocol file. It is a sample-to-result system. The ARIES HSV-1 and HSV-2 assay cassette is a disposable, single-use device that contains nucleic acid purification

reagents, an internal sample processing control and an assay-specific mastermix. Cassettes can be stored at room temperature. The systems require a universal assay protocol (i.e. identical sample preparation, amplification reagents and conditions) that can enable multiple sample types and up to 12 different IVD assays to be run together in a random batch, which is not the same as random access. Turnaround time for the HSV-1 and HSV-2 assay is approximately 2 hours. The systems are appropriate for moderate-sized laboratories.

Truelab Real Time micro PCR System (Molbio Diagnostics): Molbio Diagnostics has developed a comprehensive, rapid, near-patient real-time PCR platform called the Truelab Real Time microPCR System. The system is portable and includes all instrumentation, reagents and essential accessories that are required for the operator to conduct a real-time, quantitative PCR assay, from sample preparation through to final result reporting, all within 1 hour. A Truelab microPCR printer is also available. The system works on ready-to-use Truenat disease-specific assays that are stable at room temperature. Molbio has a large menu of assays for HCV, *C. trachomatis*, *N. gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*, *T. vaginalis* and high-risk HPV (16, 18, 31, 45).

The Truenat STI assays are:

- ***C. trachomatis***: This is an in vitro assay for the semi-quantitative detection of *C. trachomatis* in female endocervical and vaginal swab specimens, male urethral swab specimens, and male and female urine specimens as an aid in the diagnosis of symptomatic or asymptomatic infection with *C. trachomatis*. Turnaround time is 35 minutes;
- ***N. gonorrhoeae***: This is an in vitro assay for the semi-quantitative detection of *N. gonorrhoeae* in female endocervical and vaginal swab specimens, male urethral swab specimen and male and female urine specimens as an aid in the diagnosis of symptomatic or asymptomatic infection with *N. gonorrhoeae*. Turnaround time is 35 minutes;
- ***C. trachomatis/N. gonorrhoeae***: This is an in vitro assay for the semi-quantitative detection of *C. trachomatis* and *N. gonorrhoeae* in female endocervical and vaginal swab specimens, male urethral swab specimen and male and female urine specimens as an aid in the diagnosis of symptomatic or asymptomatic infection with *C. trachomatis* or *N. gonorrhoeae*. Turnaround time is 35 minutes;
- ***T. vaginalis***: This is an in vitro assay for the semi-quantitative detection of *T. vaginalis* in female endocervical and vaginal swab specimens, male urethral swab specimens, and male and female urine specimens as an aid in the diagnosis of symptomatic or asymptomatic infection with *T. vaginalis*. Turnaround time is 35 minutes;
- **High-risk HPV**: This is an in vitro assay for the semi-quantitative detection of high-risk HPV types 16, 18, 31 and 45 in clinician-collected female cervical specimens collected as an aid in the differential diagnosis of symptomatic or asymptomatic infection with high-risk HPV types 16, 31, 18 and 45. Turnaround time is 1 hour.

The testing process begins with sample collection (blood, serum or plasma) followed by extraction, which uses the Trueprep Auto Sample Prep Device and Trueprep Auto sample prep kits. The completely automated extraction process takes about 20 minutes per sample. From there, 6 µl of the extracted nucleic acid is dispensed into the reaction well of the disease-specific Truenat microPCR chip. The chip, which contains all of the chemistry required to complete an assay, is then inserted into the Truelab Uno Dx Real Time microPCR Analyzer, which is based on TaqMan chemistry. Thermal cycling takes place automatically within the analyser. The Truelab Duo and Truelab Quattro Realtime microPCR analyser systems that allow a higher throughput and random access are also commercially available.

During amplification, the Truenat microPCR chip exponentially releases fluorophores. These signals are captured by sensors and are displayed as an amplification curve on the Truelab screen. Test results are compared to lot-specific standard values pre-set into the Truenat chip, which enables quantitative estimation of the test analyte and display as real-time PCR results in approximately 30 minutes. An internal control is provided from the extraction stage for a complete validation of the test results. Test results are automatically stored in the analyser memory (up to 20 000 results), can be printed and transported wirelessly to any server or compatible device by Wi-Fi, GPRS, Bluetooth or even SMS.

SOLANA Platform, Lyra Reagents and AmpliVue (QuidelOrtho Corporation): QuidelOrtho develops and markets immunoassay and molecular diagnostic platforms and assays with a focus on POC testing. Of particular interest with regard to tests for STIs is Quidel's SOLANA platform. QuidelOrtho offers nine different molecular assays, including the SOLANA Trichomonas Assay and the SOLANA HSV-1 + HSV-2/varicella zoster virus assay, with 15 different pathogens on SOLANA, a compact benchtop instrument (24 cm × 24 cm × 15 cm). The platform features the company's proprietary helicase-dependent amplification (HDA) technology. HDA uses a helicase enzyme to unwind double-stranded DNA into single strands, eliminating the need for a thermocycler. Unlike other isothermal amplification methods, HDA uses a probe-based detection method, resulting in greater specificity. In addition, HDA permits the multiplexing of multiple pathogens in a single reaction tube.

The SOLANA platform can process individual samples or batches up to 12 samples in a single run, thus enabling cost-effective testing across a wide range of daily testing volumes. The company believes the platform is ideal for small-to-medium-sized microbiology laboratories where the low total cost of the instrument and disposables enable molecular testing at the volumes seen in these settings. The SOLANA platform is designated as moderately complex by the U.S. FDA.

The SOLANA ***T. vaginalis*** assay is a rapid IVD test for the qualitative detection of nucleic acids isolated from clinician-collected vaginal swab or urine specimens obtained from asymptomatic or symptomatic patients to aid in the diagnosis of trichomoniasis. On the SOLANA platform, a specimen is lysed by simple heat treatment, diluted and added to a reaction tube containing lyophilized HDA reagents, including primers specific for the amplification of a *T. vaginalis*-specific target sequence, as well as sequence probes. Competitive process control is included in the lysis tube to monitor sample processing, inhibitory substances in clinical samples, reagent failure or instrument failure. Results are displayed on the platform's touchscreen, can be saved to the instrument, printed and can be sent to a laboratory information system. Turnaround time for the test is 35 minutes.

The SOLANA **HSV-1** + **HSV-2**/varicella zoster virus assay is a multiplex IVD test for the detection and differentiation of HSV-1, HSV-2 and varicella zoster virus nucleic acids isolated and purified from cutaneous or mucocutaneous swab specimens obtained from symptomatic patients. The test principle is the same as that for the *T. vaginalis* assay, with the use of HSV-1, HSV-2 and varicella zoster virus-specific target sequences and fluorescence probes. Turnaround time for this assay is 50 minutes.

QuidelOrtho offers its Lyra Direct HSV-1 + HSV-2/varicella zoster virus assay. Like the SOLANA HSV-1 + HSV-2/varicella zoster virus, the Lyra Direct assay is a multiplex in vitro test that detects and differentiates HSV-1, HSV-2 and varicella zoster virus nucleic acids isolated and purified from cutaneous or mucocutaneous swab specimens obtained from symptomatic patients. It uses PCR technology and target-specific primers and fluorescently labelled probes that hybridize to conserved regions in the genomes of HSV-1, HSV-2 and varicella zoster virus. The assay has a one-step reagent set-up and a three-step sample prep. Turnaround time is less than 70 minutes. The Lyra Direct assay can be performed on the Applied Biosystems 7500 Fast Dx, QuantStudio Dx Real-Time PCR instrument and Cepheid SmartCycler II.

Finally, QuidelOrtho offers its AmpliVue HSV-1 + HSV-2 assay, which is an IVD test for the qualitative detection and differentiation of HSV-1 and HSV-2 nucleic acids isolated from the cutaneous and mucocutaneous lesions of symptomatic patients. The assay consists of three primary steps: *(1)* specimen preparation (one-step dilution); *(2)* isothermal HDA of target amplicons specific to HSV-1 and HSV-2; and *(3)* detection of the amplified DNA by target-specific hybridization probes via a colorimetric reaction (coloured lines) on a lateral flow strip that is embedded in a self-contained disposable cassette, to prevent amplicon contamination. Turnaround time is approximately 60 minutes.

EasyNAT System (Ustar Biotechnologies): Ustar Biotechnologies has developed cross priming amplification (CPA), a new isothermal molecular test with multiple iterative designs that can address a wide variety of key obstacles to traditional amplification technologies such as PCR. By using multiple crossing primers and probes, target DNA sequences can be rapidly and precisely amplified at a uniform temperature (typically 63 °C) in an easy-to-use protocol with high sensitivity and specificity. The platform is a fully integrated and automated (sample-in, answer-out) molecular diagnostic system. Turnaround time for the assays is approximately 50 minutes.

The EasyNAT system includes assays for ***N. gonorrhoeae***, ***C. trachomatis/N. gonorrhoeae*** combo test, ***T. vaginalis***, ***M. genitalium***, *U. urealyticum*, influenza A/B, respiratory syncytial virus, pertussis bacillus, HSV-1 and HSV-2, HPV 16/18 and HPV 6/11.

The *N. gonorrhoeae* DNA (CPA assay) and the *C. trachomatis*/*N. gonorrhoeae* (CPA assay) are intended for the in vitro qualitative detection of *C. trachomatis* or *N. gonorrhoeae* nucleic acid in male urinary tract swabs and female cervical swabs. The *M. genitalium* DNA (CPA assay) is intended for the in vitro qualitative detection of *M. genitalium* DNA in urine samples. The *T. vaginalis* DNA (CPA assay) is intended for the in vitro qualitative detection of *T. vaginalis* in urine and urogenital swabs. The EasyNAT HSV-1 and HSV-2 assay is intended for the in vitro qualitative detection of HSV-1 and HSV-2 in urogenital swabs. The EasyNAT HPV 6/11 assay is intended for the in vitro qualitative detection of the DNA of HPV genotypes 6 and 11 in endocervical and urogenital swabs. Similarly, the EasyNAT HPV 16/18 assay is intended for the in vitro qualitative detection of the DNA of HPV genotypes 16 and 18 in endocervical and urogenital swabs. All of the assay cartridges are designed for use on the EasyNAT platform.

The Ustar diagnostic test system consists of a reagent-containing cartridge and a portable, high-throughput device for sample preparation, amplification and detection; all are integrated and automated. Reagents consist of glassified enzymes for ambient temperature transport. With the exception of a sample extraction buffer, all reagents are preloaded and housed in the cartridge. The Ustar EasyNAT platform has been designed for both large and small health centre laboratory facilities.

Visby Medical (USA): Visby Medical (formerly Click Diagnostics) has developed a PCR-based diagnostic test that is small, portable and disposable (single-use). The test module automatically performs and integrates sample processing, RT–PCR amplification and amplicon detection without the use of separate instrumentation. Turnaround time is less than 30 minutes. The test currently requires AC mains power.

The company has developed and introduced the Visby Medical Sexual Health Click Test, which is an IVD assay for the qualitative detection of ***C. trachomatis/N. gonorrhoeae/T. vaginalis*** in women using vaginal swabs.

STI Multiplex Array/Vivalytic Analyzer (Randox Laboratories, aprimeo diagnostics, an R-Biopharm AG company): Bosch Healthcare Solutions developed the Vivalytic Analyzer, a universal, cartridge-based platform for sample-to-answer molecular diagnostics. The Vivalytic platform can accommodate a wide variety of samples and allows for different methods of analysis to run in a fully automated way in a short time frame, with results from 30 minutes. Single or multiple pathogens can be detected simultaneously in the patient sample. In addition, the Vivalytic platform is an open system that can process molecular diagnostic tests from various assay manufacturers.

The Vivalytic Analyzer has a small footprint and is a fully automated device with no peripherals, capable of quantitative and qualitative PCR procedures with three stable isothermal zones, where rapid microfluidic transfer between these zones achieves fast heating and cooling cycles. According to the company, this ensures high test quality and reproducibility. The analyser has a universal optical evaluation unit, which enables microarrays, qualitative or quantitative PCR, as well as melting curve analyses to be read out in one system. Four standard colour channels can be evaluated per PCR strand. This corresponds to a degree of multiplexing of up to eight for qualitative or quantitative PCR, or up to 16 in multichannel melting curve analysis. Via geometrical multiplexing with the help of microarrays, a much higher number can be achieved. Up to 100 properties can be examined. The Vivalytic system has built-in connectivity and can be easily integrated with popular standard IT systems. Furthermore, an analyser device can be networked and combined with many other devices, so that several series of tests can be carried out at the same time.

Randox Laboratories has developed a cartridge panel for STIs that has been adapted for use with the Vivalytic Analyzer. The STI array panel, which is CE-IVD-marked, detects 10 of the most important bacterial, viral and protozoan STIs, providing a comprehensive infection profile from a single swab sample. The test panel includes: ***C. trachomatis***, ***N. gonorrhoeae*** and ***T. vaginalis***, as well as *M. genitalium*, *U. urealyticum*, Haemophilus ducreyi, *M. hominis*, ***T. pallidum***, and **HSV-1** and **HSV-2**. The assay requires 300 µl of either a urogenital swab or urine specimen. Turnaround time is 2 hours and 30 minutes. All reagents required for a test are stored on the STI array cartridge and all are stable at room temperature; no cold storage or special shipping conditions are required. The cartridge also employs the Randox Biochip Array, a 9 × 9 mm solid-state unit that facilitates multiple target testing from a single patient sample. Each STI Biochip has 25 discrete test regions, and each discrete test region holds an individual test. A single sample is added to one cartridge, which then provides multiple test results. The cartridge contains internal controls that indicate successful extraction, amplification, hybridization and detection; all of these must pass acceptance criteria for the Vivalytic Analyzer to return patient results. Furthermore, test results do not require interpretation; positive or negative results are indicated for each target without ambiguity.

Bacterial ID systems

This section briefly introduces panels used to identify bacteria. These panels can distinguish between Gram-positive and Gram-negative bacteria, including yeast and fungi, as well as identify bacteria at the species level. Both manual and automated systems require culture specimens and can take anywhere from 2 to 48 hours. Identification is derived from a large database accessed through the company:

- *Manual enzymatic biochemical immunological tests:* BD BBL Crystal identification system, bioMérieux, API, Thermo Fisher Scientific's RapID systems;
- *Agglutination tests:* MKL Diagnostics Phadebact Monoclonal GC Test, TCS Biosciences Genochek-II Test;
- *Automated Bacterial ID systems:* Biolog microbial identification systems.

The limits of diagnostic technology

The large numbers of STIs and the variety of potential tests for each STI make the appropriate choice of diagnostic tests difficult. Tests for STIs may be used for a variety of different purposes that, in turn, may affect the choice of tests.

Despite the increasing sophistication of new diagnostic technologies, the impact of such technologies may be limited unless they can successfully accommodate the weaknesses in health care systems in resource-constrained settings, which often affect the successful delivery of diagnostics in-country. These include: shortages of human resources and lack of training for staff; supply chain challenges; lack of diagnostic equipment and equipment breakdowns; and a lack of robust quality assurance and quality control systems.

These weaknesses suggest not only the in-country need for training of test operators and service and maintenance contracts for diagnostics, but also suggest that the following operational specifications for POC diagnostic assays and platforms should be prioritized.

- **Ease of use:** Sample preparation should be simple, with the ability to use unprocessed sample specimens; only a small number of operator steps, especially timed steps, should be required to perform the test. Test kits (i.e. the reagents and disposables required to perform an assay on a single patient) should be self-contained. Specimen self-collection and possible self-testing may be a consideration with some tests to meet country and population needs.
- **Training:** The assay should be simple enough that its use can be explained to a health care worker in a day's training or less, including its methods of sample collection and preparation.
- **High tolerance to difficult environmental conditions:** Test kits must be stable at high temperature and humidity and must be able to survive extreme fluctuations in temperature; no cold chain should be required during transport and/or storage.
- **Self-contained quality control:** There should be a procedural control internalized in the cartridge for each individual test as well as an indicator of instability or test expiration.
- **Data capture, connectivity and data export:** If combined with a reader (either internal or external), the reader must store patient results and its output needs to be compatible with centralized data aggregation and analysis. To monitor test performance, a GPS/GPRS modem, preferably internal to the reader, should be incorporated, and full data export capabilities over mobile phone networks should be a minimal standard.
- **Biosafety:** To enhance biosafety, operational specifications should include the requirement for closed, self-contained systems with no biosafety cabinet required and unprocessed sample transfer only.

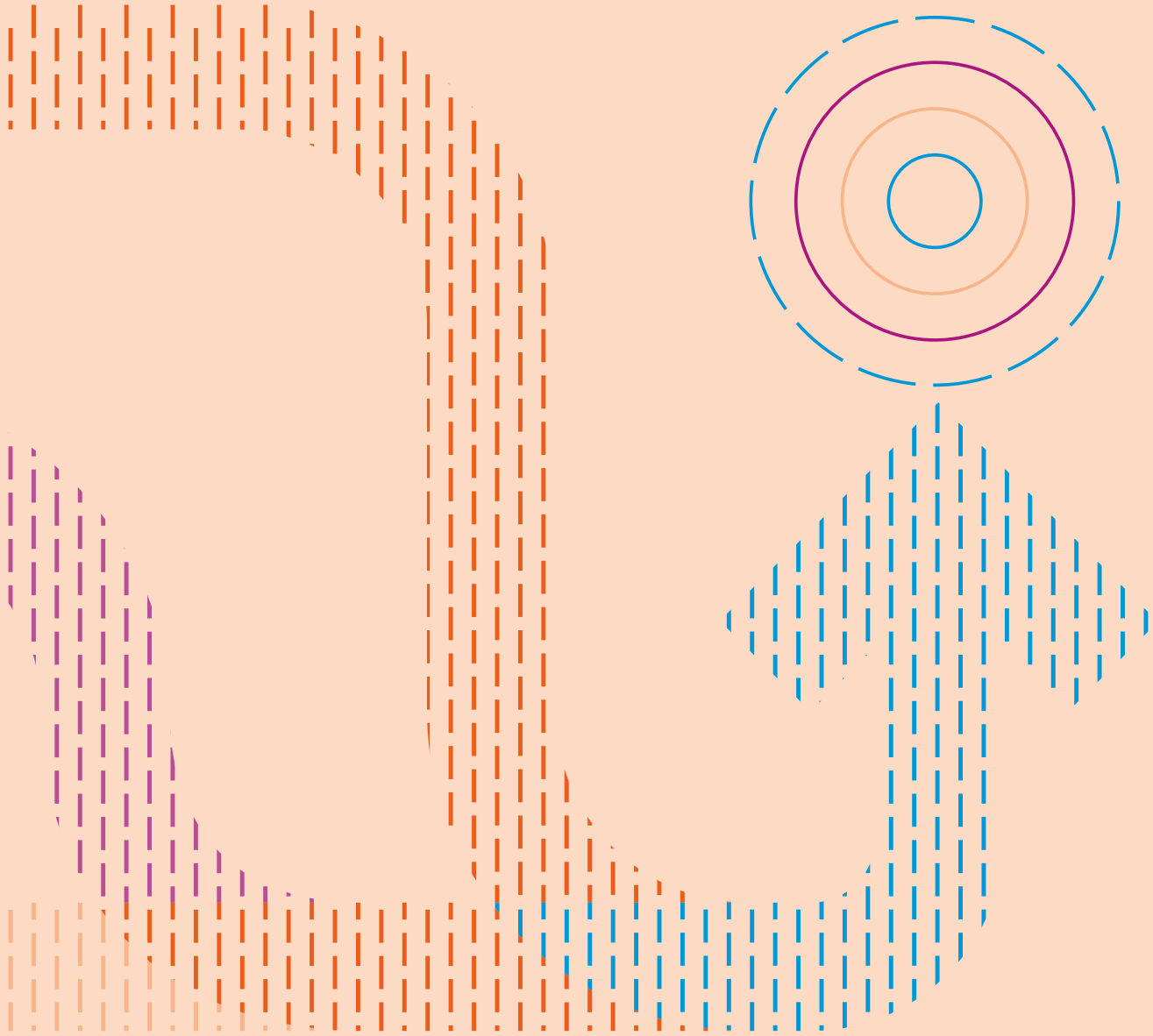
Additional high-priority specifications

In addition to the high-priority product standards summarized above, the following specifications are also important:

- **Cost:** The cost of platforms and assays will be a critical factor in the implementation and uptake of new diagnostics. Funding for diagnostics is limited, both at the global level and in-country, where cost-effectiveness will be assessed;
- **Sample capacity, throughput and time to result:** These are important specifications for new diagnostic assays, but there is no single specification for capacity, throughput and turnaround time that will fit all settings. Rather, these specifications will depend on the volume of testing and turnaround time for each assay at the target use setting (e.g. district hospital, health centre). The ability to give same-day results can be critical depending on a variety of factors (i.e. disease, morbidity, mortality) and can be considered with regard to each assay. The value of a POC test is substantially diminished if results are not provided on the same day. The working day in many health centre settings is greatly shortened (6 hours or less) and the turnaround time for a diagnosis must also allow time for the preanalytical (e.g. patient registration) and postanalytical (e.g. clinical interpretation and treatment) activities necessary to provide a complete service to the patient within one working day.

These factors, along with required technical performance, must be considered and prioritized by developers of diagnostics intended for use at or near the point of patient care in resource-limited settings.

4. Conclusions



4. Conclusions

There is a reasonably robust landscape of commercialized diagnostics for the screening and identification of syphilis, *C. trachomatis*, *N. gonorrhoeae*, *T. vaginalis*, *M. genitalium*, HPV and **HSV-1 and HSV-2** in LMICs. Most products have approval status and acceptable performance. Some lateral flow/RDT diagnostics have high specificities but low sensitivities (e.g. *C. trachomatis*, *N. gonorrhoeae*) and as such do not perform adequately to be used as screening tests; thus, improved assays are needed (18, 42, 55–57). The need is particularly acute with regard to women, where the syndromic approach to managing STIs is inadequate.

Many of the rapid diagnostic tests and molecular platforms can detect more than one STI. Arguably, the greatest need in resource-limited settings now is for a combination test for syphilis and HIV for certain target populations, including MSM, sex workers and pregnant women, could provide greater coverage for syphilis by leveraging HIV programmes. Perhaps the most acute of these needs is a dual test to help identify infections in mothers and eliminate mother-to-child transmission of HIV and syphilis, which is a significant cause of death in infants and young children globally each year *(58)*.

Furthermore, the ability to develop efficient, effective, optimized diagnostic systems is critical as resources remain limited for many global infectious diseases. Sharing of devices (by using multiplex technologies) or services and systems (i.e. sample transport, human resources, quality assurance) can improve overall diagnostics systems in countries and increase access to testing for STIs and other diseases.

In this landscape, the test characteristics for each assay have been provided and platforms identified as laboratory-based or at/near-POC, based on manufacturer information and may not be comprehensive at the time of publication. It is useful to consider a variety of assay and platform characteristics for STI programmes to effectively increase testing coverage and improve access.

While there have been evaluations for each of the assays included in this landscape, that information was not included due to the vast number of studies and lack of robust systematic reviews. The reader is encouraged to seek out this information to better understand the diagnostic accuracy and uses across various settings. Every effort was made to provide the most up-to-date information, but there will surely be updates since the release of this publication.

4.1 Limitations

This report focused on a diagnostics landscape for a multitude of different STIs. To keep the report digestible, the focus was on the characteristics and performance for commercially available approved tests appropriate for LMICs. Not all tests identified are equally accessible and appropriate across all LMIC settings, use cases and health system levels. More information is needed to link the technologies to use cases and clinical context. For example, clarity on all the different approaches to syphilis testing is needed, the pros and cons of each, where POC testing is urgently needed, to name but a few. However, the relevance of these pieces of information is likely to vary according to context and setting. WHO's updated 2023 manual, titled Laboratory and point-of-care diagnostic testing for sexually transmitted infections, including HIV, should also be referenced to provide more background and programmatic considerations.

A limitation of this report is the risk that some products or developments have been omitted due to lack of publicly available information and manufacturer nondisclosure of key product specifications. It also does not address gold standards of testing or comprehensively cover commercial product information.

5. References

1. Global progress report on HIV, viral hepatitis and sexually transmitted infections, 2021: accountability for the global health sector strategies 2016–2021: actions for impact. Geneva: World Health Organization; 2021 (https://apps.who.int/iris/handle/10665/341412, accessed 24 June 2023).
2. James C, Harfouche M, Welton NJ, Turner KM, Abu-Raddad LJ, Gottlieb SL et al. Herpes simplex virus: global infection prevalence and incidence estimates, 2016. Bull World Health Organ. 2020;98*(5)*:315–29. doi:10.2471/BLT.19237149.
3. Workowski KA, Bolan GA, Centers for Disease Control and Prevention. Sexually transmitted diseases treatment guidelines, 2015. MMWR Recomm Rep. 2015;64(RR-03):1–137.
4. Sexually transmitted infections treatment guidelines, 2021. Atlanta (GA): Centers for Disease Control and Prevention; 2021 (https://www.cdc.gov/std/treatment-guidelines/toc.htm, accessed 24 June 2023).
5. Chesson HW, Mayaud P, Aral SO. Sexually transmitted infections: impact and cost-effectiveness of prevention. In: Holmes KK, Bertozzi S, Bloom BR, Jha P, editors. Major infectious diseases, third edition. Washington (DC): The World Bank; 2017. doi:10.1596/978-1-4648-0524-0_ch10.
6. Peeling RW. Applying new technologies for diagnosing sexually transmitted infections in resource-poor settings. Sex Transm Infect. 2011;87(Suppl 2):ii28–30. doi:10.1136/sti.2010.047647.
7. Newman LR, Rowley J, Vander Hoorn S, Wijesooriya NS, Unemo M, Low N et al. Global estimates of the prevalence and incidence of four curable sexually transmitted infections in 2012 based on systematic review and global reporting. PLoS One. 2015;10*(12)*:e0143304. doi:10.1371/journal.pone.0143304.
8. Smolak A, Rowley J, Nagelkerke N, Kassebaum NJ, Chico RM, Kornromp EL et al. Trends and predictors of syphilis prevalence in the general population: global pooled analyses of 1103 prevalence measures including 136 million syphilis tests. Clin Infect Dis. 2018;66*(8)*:1184–91. doi:10.1093/cid/cix975.
9. Korenromp EL, Rowley J, Alonso M, Mello MB, Wijesooriya NS, Mahiané SG et al. Global burden of maternal and congenital syphilis and associated adverse birth outcomes – estimates for 2016 and progress since 2012. PLoS One. 2019;14*(2)*:e0211720. doi:10.1371/journal.pone.0211720.
10. Gomez GB, Kamb ML, Newman LM, Mark J, Broutet N, Hawkes SJ. Untreated maternal syphilis and adverse outcomes of pregnancy: a systematic review and meta-analysis. Bull World Health Organ. 2013;91*(3)*:217–26. doi:10.2471/BLT.12.107623.
11. Men who have sex with men. In: Global HIV, Hepatitis and STIs Programmes [website]. Geneva: World Health Organization; 2023 (https://www.who.int/teams/global-hiv-hepatitis-and-stis-programmes/populations/men-who-have-sex-with-men, accessed 24 June 2023).
12. Tsuboi M, Evans J, Davies EP, Rowley J, Korenromp EL, Clayton T et al. Prevalence of syphilis among men who have sex with men: a global systematic review and meta-analysis from 2000–20. Lancet Global Health. 2021;9*(8)*:e1110–18. doi:10.1016/S2214-109X*(21)*00221-7.
13. Syphilis – CDC detailed fact sheet. Atlanta (GA): Centers for Disease Control and Prevention; 2021 (http://www.cdc.gov/std/syphilis/stdfact-syphilis-detailed.htm, accessed 24 June 2023).
14. Sex workers with active syphilis. Geneva: Global Health Observatory (GHO) data; 2020 (https://www.who.int/data/gho/data/themes/topics/indicator-groups/indicator-group-details/GHO/sex-workers-with-active-syphilis, accessed 24 June 2023).
15. Tille PM, editor. Bailey & Scott's diagnostic microbiology, 14th edition. St. Louis (MO): Elsevier; 2017.
16. Diagnostic stewardship: a guide to implementation in antimicrobial resistance surveillance sites. Geneva: World Health Organization; 2016 (https://apps.who.int/iris/handle/10665/251553, accessed 29 June 2023).

17. Prioritization of pathogens to guide discovery, research and development of new antibiotics for drug-resistant bacterial infections, including tuberculosis. Geneva: World Health Organization; 2017 (https://apps.who.int/iris/handle/10665/311820, accessed 24 June 2023).

18. Herbst de Cortina SH, Bristow CC, Davey DJ, Klausner JD. A systematic review of point of care testing for *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, and *Trichomonas vaginalis*. Infect Dis Obstet Gynecol. 2016;2016:4386127. doi:10.1155/2016/4386127.

19. Vickerman P, Watts C, Alary M, Mabey D, Peeling RW. Sensitivity requirements for the point of care diagnosis of *Chlamydia trachomatis* and *Neisseria gonorrhoeae*. Sex Transm Infect. 2003;79*(5)*:363–7. doi:10.1136/sti.79.5.363.

20. Yang S, Zhao W, Wang H, Wang Y, Li J, Wu X. *Trichomonas vaginalis* infection-associated risk of cervical cancer: a meta-analysis. Eur J Obstet Gynecol Reprod Biol. 2018;228:166–73. doi:10.1016/j.ejogrb.2018.06.031.

21. Mielczarek E, Blaszkowska J. *Trichomonas vaginalis*: pathogenicity and potential role in human reproductive failure. Infection. 2016;44*(4)*:447–58. doi:10.1007/s15010-015-0860-0.

22. Silver BJ, Guy RJ, Kaldor JM, Jamil MS, Rumbold AR. *Trichomonas vaginalis* as a cause of perinatal morbidity: a systematic review and meta-analysis. Sex Transm Dis. 2014;41*(6)*:369–76. doi:10.1097/OLQ.0000000000000134.

23. Kissinger P, Adamski A. Trichomoniasis and HIV interactions: a review. Sex Transm Infect. 2013;89*(6)*:426–33. doi:10.1136/sextrans-2012-051005.

24. Madico G, Quinn TC, Rompalo A, McKee KT Jr, Gaydos CA. Diagnosis of *Trichomonas vaginalis* infection by PCR using vaginal swab samples. J Clin Microbiol. 1998;36*(11)*:3205–10. doi:10.1128/JCM.36.11.3205-3210.1998.

25. Wendel KA, Erbelding EJ, Gaydos CA, Rompalo AM. *Trichomonas vaginalis* polymerase chain reaction compared with standard diagnostic and therapeutic protocols for detection and treatment of vaginal trichomoniasis. Clin Infect Dis. 2002;35*(5)*:576–80. doi:10.1086/342060.

26. Advances in laboratory detection of *Trichomonas vaginalis*. Silver Spring (MD): APHL; 2013 (http://www.aphl.org/AboutAPHL/publications/Documents/ID_2013August_Advances-in-Laboratory-Detection-of-Trichomonas-vaginalis.pdf, accessed 24 June 2023).

27. Getman D, Jiang A, O'Donnell M, Cohen S. *Mycoplasma genitalium* prevalence, coinfection, and macrolide antibiotic resistance frequency in a multicenter clinical study cohort in the United States. J Clin Microbiol. 2016;54*(9)*:2278–83. doi:10.1128/JCM.01053-16.

28. Gaydos CA. *Mycoplasma genitalium*: accurate diagnosis is necessary for adequate treatment. J Infect Dis. 2017;216 (Suppl 2):S406–411. doi:10.1093/infdis/jix104.

29. Herpes simplex virus. Geneva: World Health Organization; 2023 (https://www.who.int/news-room/fact-sheets/detail/herpes-simplex-virus, accessed 24 June 2023).

30. LeGoff J, Péré H, Bélec L.Diagnosis of genital herpes simplex virus infection in the clinical laboratory. Virol J. 2014;11:83. doi:10.1186/1743-422X-11-83.

31. WHO guidelines for the treatment of genital herpes simplex virus. Geneva: World Health Organization; 2016 (https://apps.who.int/iris/handle/10665/250693, accessed 24 June 2023).

32. Genital HPV infection – basic fact sheet. Atlanta (GA): Centers for Disease Control and Prevention; 2021 (https://www.cdc.gov/std/hpv/stdfact-hpv.htm, accessed 24 June 2023).

33. Antibiotic resistance threats in the United States. Atlanta (GA): Centers for Disease Control and Prevention; 2019 (https://www.cdc.gov/drugresistance/threat-report-/pdf/ar-threats-.pdf, accessed 24 June 2023).

34. Landscape of diagnostics against antibacterial resistance, gaps and priorities. Geneva: World Health Organization; 2019 (https://apps.who.int/iris/handle/10665/326480, accessed 24 June 2023).

35. Dailey PJ, Osborn J, Ashley, EA, Baron EJ, Dance DAB, Fusco D et al. Defining system requirements for simplified blood culture to enable widespread use in resource-limited settings. Diagnostics. 2019;9*(1)*:10. doi:10.3390/diagnostics9010010.

36. Hanafiah KM, Arifin N, Bustami Y, Noordin R, Garcia M, Anderson D. Development of multiplexed infectious disease lateral flow assays: challenges and opportunities. Diagnostics. 2017;7*(3)*:51. doi:10.3390/diagnostics7030051.

37. Weile J, Knabbe C. Current applications and future trends of molecular diagnostics in clinical bacteriology. Anal Bioanal Chem. 2009;394*(3)*:731–42. doi:10.1007/s00216-009-2779-8.

38. Lin L-R, Ton M-L, Gao K, Zhen Zhu X-Z, Fan J-Y, Zheng W-H et al. A negative nontreponemal and/or specific antitreponemal IgM test does not exclude active infectious syphilis: evidence from a rabbit infectivity test: a case report. Medicine. 2016;95*(31)*:e4520. 10.1097/MD.0000000000004520.

39. Larsen SA, Steiner, BM, Rudolph AH. Laboratory diagnosis and interpretation of tests for syphilis. Clin Microbiol Rev. 1995;8*(1)*:1–21. doi:10.1128/CMR.8.1.1.

40. Luo Y, Xie Y and Xiao Y. Laboratory diagnostic tools for syphilis: current status and future prospects. Front Cell Infect Microbiol. 2021;10:574806. doi:10.3389/fcimb.2020.574806.

41. Jafari Y, Peeling RW, Shivkumar S, Claessens C, Joseph L, Pai NP. Are *Treponema pallidum* specific rapid and point-of-care tests for syphilis accurate enough for screening in resource limited settings? Evidence from a meta-analysis. PLoS One. 2013;8*(2)*:e54695. doi:101371/journal.pone.0054695.

42. Kelly H, Coltart CEM, Pant Pai N, Klausner JD, Unemo M, Toskin I et al. Systematic reviews of point-of-care tests for the diagnosis of urogenital *Chlamydia trachomatis* infections. Sex Transm Infect. 2017;93(S4):S22–30. doi:10.1136/sextrans-2016-053067.

43. Huppert JS, Mortensen JE, Reed JL, Kahn JA, Rich KD, Miller WC et al. Rapid antigen testing compares favorably with transcription-mediated amplification assay for the detection of *Trichomonas vaginalis* in young women. Clin Infect Dis. 2007;45*(2)*:194–8. doi:10.1086/518851.

44. Gaydos CA, Klausner JD, Pant Pai N, Kelly H, Coltart C, Peeling RW. Rapid and point-of-care tests for the diagnosis of *Trichomonas vaginalis* in women and men. Sex Transm Infect. 2017;93(S4):S31–35. doi:10.1136/sextrans-2016-053063.

45. Kelly H, Mayaud P, Segondy M, Pant Pai N, Peeling RW. A systematic review and meta-analysis of studies evaluating the performance of point-of-care tests for human papillomavirus screening. Sex Transm Infect. 2017;93(S4):S36–45. doi:10.1136/sextrans-2016-053070.

46. Unitaid. Cervical cancer screening and treatment of pre-cancerous lesions for secondary prevention of cervical cancer: technology landscape. Geneva: World Health Organization; 2019 (https://unitaid.org/assets/Cervical_Cancer_Technology-landscape-2019.pdf, accessed 24 June 2023).

47. Jeronimo J, Bansil P, Lim J, Peck R, Paul P, Amador JJ et al. A multicountry evaluation of *care*HPV testing, visual inspection with acetic acid, and Papanicolaou testing for the detection of cervical cancer. Int J Gynecol Cancer. 2014;24*(3)*:576–85. doi:10.1097/IGC.0000000000000084.

48. Gaydos CA, Van Der Pol B, Jett-Goheen M, Barnes M, Quinn N, Clark C et al. Performance of the Cepheid CT/NG Xpert rapid PCR test for the detection of Chlamydia trachomatics and *Neisseria gonorrhoeae*. J Clin Microbiol. 2013. doi:10.1128/JCM.03461-12.

49. Tabrizi SN, Unemo M, Golparian D, Twin J, Limnios AE, Lahra M et al. Analytical evaluation of GeneXpert CT/NG, the first genetic point-of-care assay for simultaneous detection of *Neisseria gonorrhoeae* and *Chlamydia trachomatis*. J Clin Microbiol. 2013;51*(6)*:1945–7. doi:10.1128/JCM.00806-13.

50. Garrett N, Mitchev N, Osman F, Naidoo J, Dorward J, Singh R et al. Diagnostic accuracy of the Xpert CT/NG and OSOM Trichomonas Rapid assays for point-of-care STI testing among young women in South Africa: a cross-sectional study. BMJ Open. 2019;9*(2)*:e026888. doi:10.1136/bmjopen-2018-026888.

51. Badman SG, Causer L, Guy R, Tabrizi S. Rapid laboratory assessment of a new GeneXpert molecular point-of-care test for detection of *Trichomonas vaginalis*. In: Proceedings. Australasian Sexual Health Conference, Sydney, Australia, 9–11 October 2014. (https://www.researchgate.net/publication/266319343_Rapid_laboratory_assessment_of_a_new_GeneXpert_molecular_point-of-care_test_for_detection_of_Trichomonas_vaginalis_Aust_Sex_Hlth_Conference_10_Oct_2014, accessed 25 June 2023).

52. Clifford GM, Gallus S, Herrero R, Muñoz N, Snijders PJF, Vaccarella S et al. Worldwide distribution of human papillomavirus types in cytologically normal women in the International Agency for Research on Cancer HPV prevalence surveys: a pooled analysis. Lancet. 2005;366(9490):991–8. doi:10.1016/S0140-6736*(05)*67069-9.

53. Li N, Franceschi S, Howell-Jones R, Snijders PJF, Clifford GM. Human papillomavirus type distribution in 30,848 invasive cervical cancers worldwide: variation by geographical region, histological type and year of publication. Int J Cancer. 2011;128*(4)*:927–35. doi:10.1002/ijc.25396.

54. Bristow C, Adachi K, Nielsen-Saines K, Ank B, Pilotto JH, Joao EC et al. Characteristics of the sample adequacy control (SAC) in the Cepheid Xpert CT/NG assay in female urine specimens. J Microbiol Exp. 2014;1*(4)*:00026.

55. Gaydos C, Hardick J. Point of care diagnostics for sexually transmitted infections: perspectives and advances. Expert Rev Anti Infect Ther. 2014;12*(6)*:657–72. doi:10.1586/14787210.2014.880651.

56. Huppert J, Hesse E, Gaydos CA. What is the point? How point-of-care STI tests can impact infected patients. Point Care. 2010;9*(1)*:36–46. doi:10.1097/POC.0b013e3181d2d8cc.

57. Peeling RW, Holmes KK, Mabey D, Ronald A. Rapid tests for sexually transmitted infections (STIs): the way forward. Sex Transm Infect. 2006;82(Suppl 5):v1–6. doi:10.1136/sti.2006.024265.

58. Mother-to-child transmission of HIV. In: Global HIV, Hepatitis and STIs Programmes [website]. Geneva: World Health Organization; 2023 (https://www.who.int/teams/global-hiv-hepatitis-and-stis-programmes/hiv/prevention/mother-to-child-transmission-of-hiv, accessed 24 June 2023).

Annex 1. Summary chart of laboratory-based sexually transmitted infection molecular diagnostics available

Platforms for *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*, *Trichomonas vaginalis*, *Mycoplasma genitalium*, human papillomavirus (HPV) and herpes simplex virus (HSV) types 1 (HSV-1) and 2 (HSV-2)

Platform	Technology	*C. trachomatis*	*N. gonorrhoeae*	*C. trachomatis*/*N. gonorrhoeae*	*T. vaginalis*	*M. genitalium*	HPV	HSV-1 and HSV-2
Abbott m2000 (Abbott)	Real-time PCR			√ FDA			High-risk HPV CE-IVD	
Abbott Alinity m (Abbott)	Real-time PCR			√ FDA CE-IVD	√ FDA CE-IVD	√ FDA CE-IVD	√ FDA CE-IVD	
BD ProbeTec ET (Becton Dickinson)	SDA			√ FDA CE-IVD				
BD Viper System with XTR (BD Diagnostics)	SDA and rPCR	√ FDA CE-IVD	√ FDA CE-IVD		√ FDA CE-IVD			√ FDA CE-IVD
BD Viper LT (BD Diagnostics)	SDA and rPCR	√ FDA CE-IVD	√ FDA CE-IVD				√ FDA CE-IVD	
BD COR PX/GX System (BD Diagnostics)	PCR						√ FDA CE-IVD	
BD COR PX/MX System (BD Diagnostics)	PCR			√ CE-IVD	√ CE-IVD			
BD MAX (BD Diagnostics)	PCR	√ FDA	√ FDA		√ FDA			
ExiStation Molecular Diagnostic System (Bioneer Corporation)	Real-time PCR	√	√	√		√ CE-IVD	√ CE-IVD	

Platforms for *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*, *Trichomonas vaginalis*, *Mycoplasma genitalium*, HPV and HSV-1 and HSV-2 (continued)

Platform	Technology	*C. trachomatis*	*N. gonorrhoeae*	*C. trachomatis*/*N. gonorrhoeae*	*T. vaginalis*	*M. genitalium*	HPV	HSV-1 and HSV-2
LIAISON MDX System (DiaSorin)	Real-time PCR							√ FDA
ELITe MGB InGenius Platform (ELITechGroup Solutions)	Real-time PCR	√ CE-IVD	√ CE-IVD			√ CE-IVD		
FluoroType System (Hain Lifescience)	qPCR	√ CE-IVD	√ CE-IVD					
Hologic Panther (Hologic)	HPA			√ FDA CE-IVD	√ FDA CE-IVD	√ FDA CE-IVD	√ FDA CE-IVD	√ FDA CE-IVD
Cervista HTA System (Hologic)	iNAAT						√ FDA	
Alethia Molecular Diagnostic System (Meridian Bioscience)	LAMP							√ FDA
QIAsymphony SP/AS and Rotor-Gene Thermocyclers (QIAGEN)	Real-time PCR			√ CE-IVD				√ CE-IVD
diagene Hybrid Capture 2 and Rapid Capture System (QIAGEN)	Hybridization and chemiluminescence	√ FDA	√ FDA	√ FDA			√ FDA	

Platforms for *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*, *Trichomonas vaginalis*, *Mycoplasma genitalium*, HPV and HSV-1 and HSV-2 (continued)

Platform	Technology	*C. trachomatis*	*N. gonorrhoeae*	*C. trachomatis*/*N. gonorrhoeae*	*T. vaginalis*	*M. genitalium*	HPV	HSV-1 and HSV-2
NeuMoDx Molecular System (QIAGEN)	Real-time PCR			√ CE-IVD	√ CE-IVD	√ CE-IVD	√ CE-IVD	
cobas 6800/cobas 8800 (Roche)	Real-time PCR			√ FDA CE-IVD	√ FDA CE-IVD	√	√ FDA CE-IVD	
cobas 4800 (Roche)	Real-time PCR			√ FDA CE-IVD			√ FDA CE-IVD	√ FDA CE-IVD

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; FDA: U.S. Food and Drug Administration; HPA: hybridization protection assay; iNAAT: isothermal nucleic acid amplification technology; LAMP: loop-mediated isothermal amplification; PCR: polymerase chain reaction; qPCR: quantitative PCR; random PCR: SDA: strand displacement amplification.

Annex 2. Summary chart of sexually transmitted infection point-of-care (POC) and near-POC molecular diagnostics available

Platforms for *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*, *Trichomonas vaginalis*, *Mycoplasma genitalium*, human papillomavirus (HPV) and herpes simplex virus (HSV) types 1 (HSV-1) and 2 (HSV-2)

Platform	Technology	*C. trachomatis*	*N. gonorrhoeae*	*C. trachomatis*/*N. gonorrhoeae*	*T. vaginalis*	*M. genitalium*	HPV	HSV-1 and HSV-2
Available platforms/assays								
GeneXpert Cepheid	PCR-based NAAT	N/A	N/A	√ CE-IVD FDA	√ CE-IVD FDA	N/A	√ CE-IVD	N/A
Solana QuidelOrtho	iNAAT-HDA	N/A	N/A	N/A	√ FDA CE-IVD	N/A	N/A	√ HIV-1 and HIV-2/ varicella zoster virus CE-IVD FDA
***care*HPV System** QIAGEN	Nucleic acid hybridization	N/A	N/A	N/A	N/A	N/A	√ CE-IVD	N/A
Truelab RT micro PCR Molbio	Real-time PCR	√ CE-IVD	√ CE-IVD	√ CE-IVD	√ CE-IVD	N/A	√ CE-IVD	N/A
***io* Diagnostic System** binx health	PCR-based NAAT; electrochemical detection	N/A	N/A	√ FDA CE-IVD			N/A	N/A
EasyNAT Ustar	iNAAT – CPA	N/A	√ CE-IVD	√	√ CE-IVD	√ CE-IVD	√ CE-IVD	√ CE-IVD
Visby Medical	PCR-based NAAT	N/A	N/A	√ CT/NG/TV FDA	√	N/A	N/A	N/A
HG Swift HiberGene Diagnostics	Isothermal LAMP; Fluorometric detection	N/A	N/A	√ CE-IVD	N/A	√ CE-IVD	N/A	√ CE-IVD

Platforms for *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, *C. trachomatis*/*N. gonorrhoeae*, *Trichomonas vaginalis*, *Mycoplasma genitalium*, HPV and HSV-1 and 2 HSV-2 (continued)

Platform	Technology	*C. trachomatis*	*N. gonorrhoeae*	*C. trachomatis*/*N. gonorrhoeae*	*T. vaginalis*	*M. genitalium*	HPV	HSV-1 and HSV-2
ARIES and ARIES M1 Luminex	Real-time PCR	N/A	N/A	N/A	N/A	N/A	N/A	√ FDA CE-IVD
GenomeEra CDX Abacus Diagnostica	PCR or real-time PCR; fluorescence	N/A	N/A	N/A	N/A	N/A	N/A	√ HSV 1&2, varicella zoster virus + EV CE-IVD marked
Genie II & Genie III **eazyplex** Amplex/OptiGene	iNAAT; fluorescence	√ CE-IVD		√ Plus *C. trachomatis*, *N. gonorrhoeae*, *U. urealyticum*, *M. hominis*, *M. genitalium* and *T. pallidum* combo test CE-IVD	√	√		iNAAT; fluorescence
Vivalytic Randox/Bosch	PCR-based NAAT	N/A	N/A	√ *C. trachomatis*/*N. gonorrhoeae*/*T. vaginalis*/*M. genitalium*/HSV-1 and HSV-2, plus CE-IVD	√		N/A	√ CE-IVD

CE-IVD: Conformité Européenne (CE)-marked in vitro diagnostic medical device; CPA: cross priming amplification; FDA: U.S. Food and Drug Administration; HDA: helicase-dependent amplification; HPA: hybridization protection assay; iNAAT: isothermal nucleic acid amplification test; LAMP: loop-mediated isothermal amplification; NAAT: nucleic acid amplification test; PCR: polymerase chain reaction.

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