visby medical™

IVD Rx Only

Sexual Health

Instructions for Use

REF PS-001402

Device Name

Visby Medical Sexual Health Test

Common or Usual Name

Visby Sexual Health Test

CLIA Waived

A Certificate of Waiver is required to perform this test in a CLIA Waived setting. To obtain CLIA waiver information and a Certificate of Waiver, please contact your state health department. Additional CLIA waiver information is available at the Centers for Medicare and Medicaid website at www.cms.hhs.gov/CLIA.

Failure to follow the instructions or any modifications to the test will result in the test no longer meeting the requirements for waived classification.

Intended Use

The Visby Medical Sexual Health Test is a single-use (disposable), fully integrated, automated Polymerase Chain Reaction (PCR) in vitro diagnostic test intended for use in point-of-care or clinical laboratory settings for the rapid detection and differentiation of DNA from *Chlamydia trachomatis*, *Neisseria gonorrhoeae*, and *Trichomonas vaginalis* in self-collected female vaginal swab specimens using the Visby Medical Sexual Health Vaginal Specimen Collection Kit in a health care setting. The test results are to aid in the diagnosis of symptomatic or asymptomatic infections with *Chlamydia trachomatis, Neisseria gonorrhoeae*, and *Trichomonas vaginalis*.

Summary and Explanation of the Procedure

Chlamydia trachomatis (CT), *Neisseria gonorrhoeae* (NG), and *Trichomonas vaginalis* (TV) are three of the most common Sexually Transmitted Infections (STIs) in the United States. These infections are called "silent infections" as many infected people are asymptomatic and lack abnormal physical examination findings. In 2018, the Centers for Disease Control and Prevention (CDC) estimated that 4 million chlamydial infections, 1.6 million gonococcal infections, and over 2 million trichomoniasis infections occur annually in the United States but more than half are not reported because most people are asymptomatic and are therefore not tested.[1,2,3]

CT are gram-negative, nonmotile obligate intracellular bacteria. This species currently includes nineteen serovars that can induce disease in humans, with Serovars D through K known to be the major cause of genital chlamydial infections in men and women.[4] If CT infections are left untreated, they can cause non-gonococcal urethritis, acute salpingitis (an infection of the fallopian tubes), proctitis, cervicitis, and epididymitis. Furthermore, in women, untreated CT can cause pelvic inflammatory disease (PID) in about 10-15% of the infected population which can subsequently cause permanent damage to fallopian tubes, uterus, or surrounding tissues which can result in chronic pelvic pain, infertility, and ectopic pregnancy.[2]

NG, the causative agent of gonorrheal disease, are gram-negative, non-motile diplococci. Although most Gonococcal infections are uncomplicated lower and upper genital tract infections, some can also be asymptomatic. Testing is essential in these cases as untreated NG infections in women can lead to PID which can then cause the onset of salpingitis, pelvic peritonitis, tuboovarian abscesses, and endometritis. Untreated NG infections may also develop Disseminated Gonococcal Infection (DGI),

a potentially life threating condition which is usually characterized by arthritis, tenosynovitis and/or dermatitis.[1]

TV is a protozoan parasite that causes trichomoniasis, the most common curable STI, with approximately 2 million new cases occurring annually in the US. However, only approximately 30% of TV infections are symptomatic.[3] Women who are symptomatic may experience vaginal discharge, vulvovaginal soreness, dysuria and/or irritation. Patients with trichomoniasis have an increased risk of getting and spreading other sexually transmitted infections, including HIV. In addition, pregnant women with trichomoniasis are at higher risk of premature labor and having low-birth-weight babies.[3]

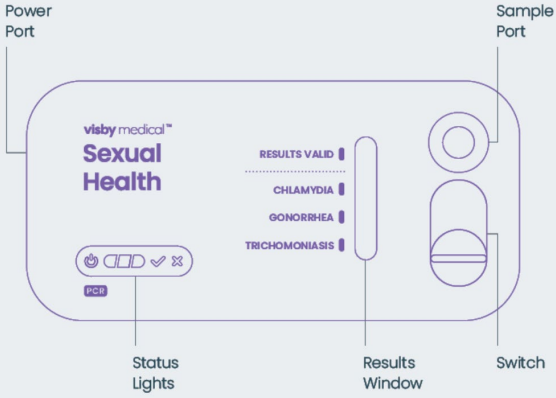
Principles of the Procedure

The Visby Medical Sexual Health Test is a single-use (disposable), fully integrated, rapid, compact device containing a PCR-based assay for direct qualitative detection and differentiation of DNA from CT, NG, and TV. The test system includes the Visby Medical Sexual Health device, the Visby Medical power supply, the Visby Medical Vaginal Specimen Collection kit, and fixed-volume transfer pipettes. The device processes a vaginal swab sample by automatically performing all steps required to complete lysis, polymerase chain reaction, and amplicon detection.

The patient uses the Visby Medical Vaginal Specimen Collection Kit to self-collect a vaginal specimen with the provided flocked swab, and then the patient elutes the specimen into the Visby Medical Collection Media. The test operator transfers the collection media containing the patient specimen into the sample port of the device using the provided fixed-volume pipette where it rehydrates a lyophilized internal process control. The sample enters a lysis module, where the DNA of the sample and the internal process control are extracted using a combination of chemical lysis and high temperature. The extracted DNA enters a mixing chamber where it rehydrates lyophilized PCR reagents, followed by thermocycling to amplify target DNA. If present, the amplified pathogen target (CT, NG, and/or TV) and internal process control hybridize to specific probes located on a flow channel. Detection of the target-specific PCR product is accomplished via an enzyme-linked colorimetric assay using streptavidin bound horseradish peroxidase (HRP) and a colorimetric substrate that forms a purple precipitate. Test results can be expected in approximately 30 minutes: a green check mark will appear, and a purple color will appear in the “Results Valid” spot, indicating a valid test. A purple spot adjacent to “Chlamydia,” “Gonorrhea,” and/or “Trichomoniasis” signifies the presence of amplified CT, NG, and/or TV DNA in the sample.

Materials Required

Materials Provided in 10 Pack Test Kit

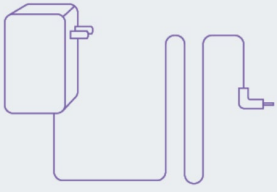


Visby Medical Sexual Health Test– Quantity 10

Visby Fixed Volume Pipette (650 µl) – Quantity 12

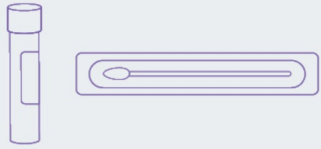
Biohazard Bag – Quantity 12

Materials Required and Available as Accessories



Visby Power Adapter

(PS-000885 or PS-000288)

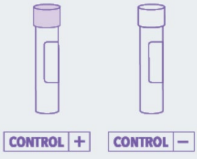


Visby Medical Sexual Health Vaginal Specimen Collection Kit‡

Materials Required but Not Provided

Gloves

Disposal Bin



External Controls

NATtrol™ *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae*(NG), *Trichomonas vaginalis*(TV) by ZeptoMetrix Corporation

\+ Control SKU:

NATCTNGTV-POS-IVD

\- Control SKU:

NATCTNGTV-NEG-IVD

‡**Note:** The Visby Medical Sexual Health device can only be used with the Visby Medical Sexual Health Vaginal Specimen Collection Kit.

Warnings and Precautions

General

1. For in vitro diagnostic use. Rx only.
2. For use only with female vaginal swabs collected in the Visby Medical Sexual Health Vaginal Specimen Collection Kit.
3. Do not re-sterilize unused swabs from the Visby Medical Sexual Health Vaginal Specimen Collection Kit.
4. This product is for single use only; do not reuse the Visby Medical Sexual Health device.
5. Federal law restricts the sale of this device unless it is by or on the order of a licensed practitioner (US only).
6. Colorblind users may be unable to differentiate red, green, and white status lights. However, they can consult the light location and shape of the light to determine test status. When interpreting results, the purple shade may appear as a dark shade for some users.
7. Results from The Visby Medical Sexual Health device must be interpreted in accordance with these Instructions for Use.
8. The specimen collection media is a clear colorless, and odorless solution. Do not use if the solution appears discolored or has a strong odor.

Safety and Contamination Prevention

1. Follow your institution's safety procedures for working with chemicals and handling biological samples.
2. Wear gloves while handling samples. If the gloves come in contact with specimen or appear to be wet, change gloves to avoid contaminating other specimens. Change gloves between processing of each specimen and before leaving the work area and upon entry into work areas.
3. Keep the work area clean to prevent contamination.
4. Do not try to disassemble the Visby Medical Sexual Health device.
5. Treat all biological specimens, including a used Visby Medical Sexual Health device, as if capable of transmitting infectious agents. All biological specimens should be treated with standard precautions. Guidelines for specimen handling are available from the U.S. Centers for Disease Control and Prevention and the Clinical Laboratory Standards Institute.[5,6]
6. If a spill with the Visby Medical Sexual Health Test occurs, soak up spill with an absorbent material. Spray the contaminated area and materials with 10% bleach. Wipe down the surface so that it is saturated with bleach and let rest for at least 10 minutes. Once a minimum of 10 minutes has passed, wipe the area down with an absorbent material, such as paper towels, followed by rinsing the area with water. Discard the Visby Medical Sexual Health device according to your Institution's standard practices.
7. If a spill occurs on the Visby power adapter, unplug the unit and wipe it down vigorously with 70% ethyl or isopropyl alcohol. Allow the power adapter to completely dry before using it again.
8. Safety Data Sheets (SDS) are available at Visby Medical Customer Support 1-833-GoVisby (1-833-468-4729).

Electromagnetic Compatibility (EMC) Safety

1. Use of this equipment adjacent to or stacked with other equipment should be avoided because it could result in improper operation. If such use is necessary, this equipment and the other equipment should be observed to verify that they are operating normally.

2. Portable RF communications equipment (including peripherals such as antenna cables and external antennas) should be used no closer than 30 cm (12 inches) to any part of the Visby Medical Sexual Health Test, including cables specified by the manufacturer. Otherwise, degradation of the performance of this equipment could result.

Visby Medical Sexual Health Test and Accessories

1. Do not use the Visby Medical Sexual Health device if it has been dropped or appears to be broken or is past its expiration date.
2. Do not shake or tilt the Visby Medical Sexual Health device after adding a sample.
3. Store the Visby Medical Sexual Health device sealed in the foil pouch prior to use.
4. Do not move or unplug the Visby Medical Sexual Health device while it is running.
5. The Visby power adapter should be replaced if an increased number of RED X errors are observed. Failure to do so may result in invalid results.
6. Only use the supplied Visby power adapter (9 V, 3.5 A DC) to power the Visby Medical Sexual Health device. Using other power adapters to operate the Visby Medical Sexual Health device will void the safety protection of the device and could result in increased electromagnetic emissions or decreased electromagnetic immunity of this equipment and result in improper operation.
7. The Visby Medical Sexual Health device is best used in a room with adequate lighting and away from glare. Failure to do so may result in an inability to see the results on the test.
8. After use, the Visby Medical Sexual Health device should be placed in a Biohazard Bag prior to disposal.
9. The Visby Medical Sexual Health device should be disposed of in the appropriate specimen waste containers according to the Institution's standard practices.
10. The results of the Visby Medical Sexual Health device must be read within 120 minutes (2 hours) after the green check mark light appears.
11. The purple switch on the Visby Sexual Health device must be fully closed to start the test. Failure to completely close the switch will result in failure to start the test and can cause invalid test results.
12. The Visby Sexual Health device should be placed and operated on a flat surface with the front of the device facing up. Placing the device at 90ºC on its side can result in invalid or false negative test results.

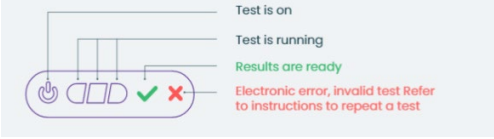
Note: Dispose of the power adapter per your local, federal, and institutional guidelines.

Specimen

1. Only patient-collected vaginal swab specimens taken with the Visby Medical Sexual Health Vaginal Specimen Collection Kit should be used with The Visby Medical Sexual Health Test.
2. Failure to add sufficient sample volume to the test can lead to invalid results.
3. The Visby Medical Sexual Health device requires a specific volume of specimen. Use the provided fixed-volume pipette to transfer sample to the device.
4. The Visby Medical Sexual Health Test should not be used if antiperspirants and deodorants or the following vaginal products: douches, washes, lubricants, vaginal wipes, vaginal moisturizers, or feminine hygiene spray in the genital area, were used by the patient within 48 hours of collection.
5. Do not dilute patient samples.
6. Samples can contain inhibitors that may generate invalid results.

Color Blindness Precaution

While colorblind users may be unable to differentiate red, green, and white status lights, they can consult the light location and shape of the light to determine test status.



Test Kit and Device Storage

Store the Visby Medical Sexual Health Test kit and device between 36°F and 86°F (2°C and 30°C), and between 5% and 80% humidity. Do not freeze. In case of refrigeration or other exposure to cold temperatures, ensure that the Visby Medical Sexual Health device is allowed to fully come to at least its minimum operating temperature of 55°F (13°C) prior to use.

Specimen Collection and Storage

Female vaginal swab specimens tested with the Visby Medical Sexual Health Test must be collected by the patient in clinical settings in accordance with the Visby Medical Sexual Health Vaginal Specimen Collection Kit Instructions for Use and following the Self-Collection Instructions. The performance of this test has not been evaluated with other specimen types or other specimen collection devices.

Note: Ensure that patients read and understand the Self-Collection Instructions before providing them with the Visby Medical Sexual Health Vaginal Specimen Collection Kit.

The Visby Medical Sexual Health Vaginal Specimen Collection Kit may contain irritants. Do not ingest the contents of the tube. If the contents of the tube are splashed in your eyes, flush your eyes with water. If the contents splash onto your skin, wash with soap and water. If irritation persists, notify a health care provider.

For best results, patient-collected samples should be tested as soon as possible. If the sample cannot be immediately tested, they can be stored as shown in the following table.

Patient Sample Storage Specifications

Room Temperature	Refrigerated	Frozen
Up to 4 hours between **64°F - 86°F** (18°C - 30°C)	Up to 4 hours between **36°F - 46°F** (2°C – 8°C)	Up to 90 days at less than **5°F** (<-15°C)

For refrigerated specimens, ensure the cap is securely tightened. Invert specimen tube at least 5 times to re-suspend any settled specimen particulates or mucus before loading into the Visby Medical Sexual Health device.

For frozen specimens, allow the tube to reach room temperature for 1-2 hours prior to use. Once at room temperature, ensure the cap is securely tightened. Invert specimen tube at least 5 times to re-suspend any settled specimen particulates or mucus before transferring he specimen into the Visby Medical Sexual Health device.

Visby Medical Sexual Health Test Procedure

Follow these instructions carefully. This test is designed for use by health care professionals.

Operating Conditions

TEMPERATURE	HUMIDITY	PRESSURE
55°F - 91°F (13°C - 33°C)	**5% - 80%**	**-300ft – 9500ft** (102.5 kPa – 71 kPa)

Run the test on a clean, level surface. Test samples within four hours of collection.

Setup | Prepare the Workspace Place device on a level surface

A. Clean your workspace and gather all the required materials, then **unwrap** the device.
Note: Use the device **immediately** after unwrapping. Use **new** gloves for each test.

B. Plug the **Visby Power Adaptor** into the device power port.

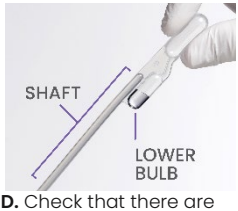
C. Remove the **sticker** over the sample port.

Step 1 | Mix and Add Patient Sample

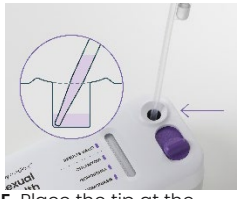
A. Mix the patient sample (or control) by **gently inverting** the tube **5 times**.

B. Squeeze the **upper bulb** of the provided pipette and submerge the tip to the **bottom** of the sample tube.

C. Release the upper bulb **slowly** to fill the shaft. Keep pipette tip submerged until shaft is full. Extra fluid should enter the lower bulb.



D. Check that there are **no air bubbles** in the shaft. **Note:** Do not squeeze lower bulb or invert the pipette.

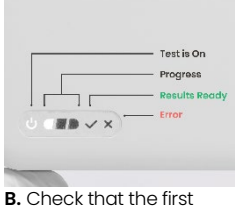


E. Place the tip at the **bottom** of the sample port and then squeeze the **upper bulb** of the pipette to release **all** of the sample.

F. Discard the pipette according to your institution's guidelines immediately. **Do not set it down.**

Step 2 | Run the Test Do not move test while running

A. Immediately after sample addition, slide the **switch upwards** in a firm, swift motion to **fully** close the sample port to ensure the test is started.

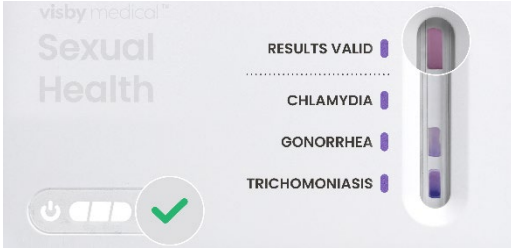


B. Check that the first progress **indicator light is blinking**. Lights will initially blink and then become stable as the test progresses.

C. Wait approximately **28 minutes** for a green check mark to appear indicating the test is finished running. **Note:** If a **red X** appears at any point, stop, do not read test results, and retest.

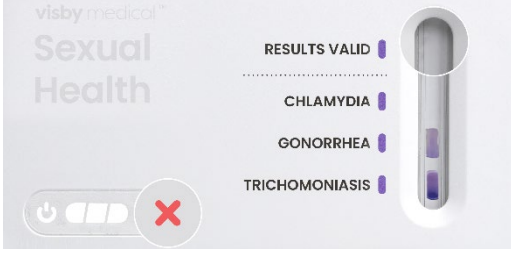
Step 3 | Interpret Results If the test is invalid, stop, and retest!

A. Determine if the test is valid.



Green check and purple spot next to "Results Valid" = **Valid Test**

Read Results



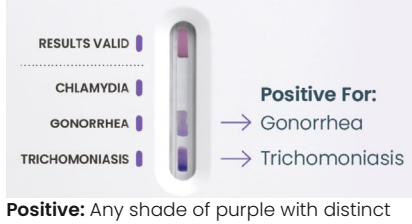
Red X or no purple spot next to "Results Valid" = **Invalid Test**

Stop and Retest

B. Read the test results. Note: Results may be read up to 2 hours after the test is completed. Do no read results if test is invalid.

Examples of possible results:

Negative: No spot next to the target is a negative result.



Positive: Any shade of purple with distinct edges next to the target is a positive result.

Intensity of the spot may vary. Any shade of color with distinct edges should be considered positive.

Interpretation of Results: Use the following table to determine next steps and reporting of patient test results.

Indicator Light	Results Valid Spot	Chlamydia Spot	Gonorrhea Spot	Trichomoniasis Spot	Next Steps	Test Results
Red X	N/A	N/A	N/A	N/A	Retest	Invalid
Green ✓	Absent	N/A	N/A	N/A	Retest	Invalid
Green ✓	Present	Present	Absent	Absent	Report Results	***Chlamydia trachomatis*** **Detected** *Neisseria gonorrhoeae* Not Detected *Trichomonas vaginalis* Not Detected
Green ✓	Present	Absent	Present	Absent	Report Results	*Chlamydia trachomatis* Not Detected ***Neisseria gonorrhoeae*** **Detected** *Trichomonas vaginalis* Not Detected
Green ✓	Present	Absent	Absent	Present	Report Results	*Chlamydia trachomatis* Not Detected *Neisseria gonorrhoeae* Not Detected ***Trichomonas vaginalis*** **Detected**
Green ✓	Present	Present	Present	Absent	Report Results	***Chlamydia trachomatis*** **Detected** ***Neisseria gonorrhoeae*** **Detected** *Trichomonas vaginalis* Not Detected
Green ✓	Present	Present	Absent	Present	Report Results	***Chlamydia trachomatis*** **Detected** *Neisseria gonorrhoeae* Not Detected ***Trichomonas vaginalis*** **Detected**
Green ✓	Present	Absent	Present	Present	Report Results	*Chlamydia trachomatis* Not Detected ***Neisseria gonorrhoeae*** **Detected** ***Trichomonas vaginalis*** **Detected**
Green ✓	Present	Present	Present	Present	Report Results	***Chlamydia trachomatis*** **Detected** ***Neisseria gonorrhoeae*** **Detected** ***Trichomonas vaginalis*** **Detected**
Green ✓	Present	Absent	Absent	Absent	Report Results	*Chlamydia trachomatis* Not Detected *Neisseria gonorrhoeae* Not Detected *Trichomonas vaginalis* Not Detected

Retest Procedure

If a retest is required, obtain the leftover sample from the Visby collection media tube. If the leftover sample has been stored for ≤ 4 hours, then repeat the test with a new Visby Medical Sexual Health device. If the leftover sample has exceeded the storage recommendations (four hours at room temperature or under refrigeration), and/or if the sample volume is insufficient, collect a new sample and repeat the test with a new Visby Medical Sexual Health Test. If a retest continues to return an invalid result, collect a new sample and repeat the test with a new Visby Medical Sexual Health Test.

If the positive or negative external controls fail, repeat the test with a new Visby Medical Sexual Health device.

If a repeat test fails, please contact Visby Medical Customer Support at 1-833-468-4729 (1-833-GoVisby).

Quality Control

The Visby Medical Sexual Health Test has built-in procedural controls. These include an internal process control and built-in electronic control. The result of the process control is displayed in the detection window while the results of the electronic controls are displayed using the status lights.

Internal Process Control

The Visby Medical Sexual Health device contains an internal process control. The internal process control monitors effective sample preparation, PCR amplification, and detection. If these steps are completed successfully, then a purple spot will develop next "Results Valid" in the detection window. If the purple spot does not appear, the test result is Invalid, and the test must be repeated with a new Visby Sexual Health device.

Built-in Electronic Control

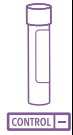
The electronic controls monitor the device to ensure proper operation. If the electronic control passes, a green check mark status light appears. If this control fails, a "Red X" status light appears. A "Red X" error will occur if (1) the device is being operated outside of its temperature range, (2) the power is interrupted during a run, (3) a device error is detected, or (4) the test is not started within 2 hours of the device being plugged in. In the event of a "Red X", the test is invalid, and the test must be repeated with a new Visby Sexual Health device.

External Positive and Negative Controls

Good laboratory practice suggests the use of positive and negative controls to ensure that the test is working properly and that the test is correctly performed.

Test these controls using the same process that is used for testing a patient sample. The external control should be run once with each new shipment of test kits and once for each untrained operator. Further controls may be tested to conform with local, state and/or federal regulations, accrediting groups, or your laboratory's standard Quality Control procedures.

NATtrol™ *Chlamydia trachomatis* (CT), *Neisseria gonorrhoeae* (NG), *Trichomonas vaginalis* (TV) Controls by ZeptoMetrix™ Corporation

Product Code	Unit	Control Key
NATtrol™ CT/NG/TV Positive Control SKU: NATCTNGTV-POS-IVD	Six (6) 1 mL Vials per Kit 	Valid Positive Control Run 
NATtrol™ CT/NG/TV Negative Control SKU: NATCTNGTV-NEG-IVD	Six (6) 1 mL Vials per Kit 	Valid Negative Control Run 

Limitations

1. The performance characteristics of the Visby Medical Sexual Health Test have not been evaluated in women less than 14 years of age.
2. This test has only been validated with female vaginal specimens self-collected in clinical settings, using the Visby Medical Sexual Health Vaginal Specimen Collection Kit. The performance of this test has not been evaluated with other specimen types or other specimen collection devices.
3. Not for use with self-collected specimens in home setting.
4. Erroneous results may occur from improper specimen collection, technical error, sample mix-up, or if the organism in the patient sample is below the limit of detection of the Visby Medical Sexual Health Test.
5. Careful compliance with the instructions in this insert, the Quick Reference Guide Instructions, and Visby Medical Sexual Health Vaginal Specimen Collection Kit instruction documents are necessary to avoid erroneous results.
6. Because the detection of CT, NG, and TV are dependent on the DNA present in the sample, reliable results are dependent on proper sample collection, handling, and storage.
7. As with other assays of this type, there is a risk of false negative or invalid results due to the presence of sequence variants (mutations) in the amplification targets.
8. A negative result does not preclude a possible infection. If clinical symptoms persist, additional testing should be performed.

9. The Visby Medical Health Test should not be used by patients using antiperspirants and deodorants or the following vaginal products: douches, washes, lubricants, vaginal swabs, vaginal moisturizers, or feminine hygiene spread in the genital area, within 48 hours of sample collection.
10. This test has been evaluated with human specimen material only.
11. The effect of interfering substances has been evaluated only for those listed in the interfering substances section of this document.
12. Assay interference was observed in the presence of the following substances: RepHresh Odor Eliminating pH Balancing Gel at a concentration greater that 1.25% w/v; Replens Long Lasting Vaginal Moisturizer at a concentration greater that 2.5% w/v; Dove 0% alcohol antiperspirant spray at concentration greater than 0.19% w/v.
13. Results from the Visby Sexual Health Test should be interpreted in conjunction with other clinical and laboratory data available to the clinician.
14. In cases where it is suspected the reporting of a positive result will have adverse socioeconomic impact, a new sample should be collected, and tested with an alternate assay.
15. The Visby Sexual Health Test is not intended for the evaluation of suspected sexual abuse or for other medico-legal indications.

Equivalence between the Visby Medical Sexual Health Test and the Visby Medical Sexual Health Click Test

The Visby Medical Sexual Health Test (Ref #: PS-001402, referenced as Visby Test hereafter) is an updated version of original test, the Visby Medical Sexual Health Click Test (Ref #: PS-000175, referenced as Click Test hereafter). Both tests use the same reagents and are composed of the same materials. The housing and fluidic path of the Visby Test was modified to improve test usability and reliability; however, the test was developed to be functionally equivalent to the original test. Clinical performance presented in the following section, was established based on the data from the Click Test. For the purpose of demonstrating that the performance of the Visby Test is equivalent to the original version of the test, clinical comparison studies were performed. The data is shown in the “Clinical Comparison Study between Visby Test and Click Test” section of this document.

Expected Values

The prevalence of infection with CT, NG, and/or TV in patient populations depends on risk factors such as age, gender, the presence or absence of symptoms, the type of clinic, and the sensitivity of the test used to detect infections. During the clinical evaluation of the Click Test, the positivity rate for detection of CT, NG, and/or TV using the Click Test, by clinical study site and overall, is shown in the table below. These values also apply to the Visby Test.

Table 1. Positivity Rate of the Visby Test for Detection of CT, NG, and/or TV during the Clinical Study

Site	% Percent Positive (# positive / # tested)						
	CT only positive	NG only positive	TV only positive	CT & NG positive	CT & TV positive	NG & TV positive	CT, NG, & TV positive
1	2.7% (5/185)	0.5% (1/185)	1.1% (2/185)	0.0% (0/185)	0.0% (0/185)	0.0% (0/185)	0.0% (0/185)
2	2.1% (5/236)	1.3% (3/236)	18.6% (44/236)	0.0% (0/236)	2.5% (6/236)	0.0% (0/236)	0.0% (0/236)
3	1.5% (1/67)	1.5% (1/67)	9.0% (6/67)	0.0% (0/67)	0.0% (0/67)	0.0% (0/67)	0.0% (0/67)
4	21.2% (57/269)	4.1% (11/269)	8.2% (22/269)	3.3% (9/269)	3.7% (10/269)	1.1% (3/269)	2.2% (6/269)
5	1.6% (1/61)	1.6% (1/61)	14.8% (9/61)	0.0% (0/61)	1.6% (1/61)	1.6% (1/61)	3.3% (2/61)
6	8.6% (23/269)	0.7% (2/269)	0.4% (1/269)	0.4% (1/269)	0.4% (1/269)	0.0% (0/269)	0.0% (0/269)
7	3.0% (10/332)	0.6% (2/332)	12.3% (41/332)	0.3% (1/332)	1.2% (4/332)	0.0% (0/332)	0.3% (1/332)
8	0.0% (0/15)	6.7% (1/15)	13.3% (2/15)	0.0% (0/15)	0.0% (0/15)	0.0% (0/15)	0.0% (0/15)
9	5.1% (3/59)	0.0% (0/59)	10.2% (6/59)	1.7% (1/59)	1.7% (1/59)	0.0% (0/59)	0.0% (0/59)
10	3.9% (2/51)	0.0% (0/51)	7.8% (4/51)	0.0% (0/51)	2.0% (1/51)	0.0% (0/51)	2.0% (1/51)
11	6.4% (5/78)	1.3% (1/78)	5.1% (4/78)	1.3% (1/78)	0.0% (0/78)	0.0% (0/78)	0.0% (0/78)
12	10.5% (6/57)	0.0% (0/57)	5.3% (3/57)	5.3% (3/57)	0.0% (0/57)	1.8% (1/57)	0.0% (0/57)
13	12.7% (8/63)	1.6% (1/63)	3.2% (2/63)	1.6% (1/63)	3.2% (2/63)	1.6% (1/63)	3.2% (2/63)
14	10.6% (5/47)	2.1% (1/47)	6.4% (3/47)	0.0% (0/47)	2.1% (1/47)	0.0% (0/47)	0.0% (0/47)
All	7.3% (131/1789)	1.4% (25/1789)	8.3% (149/1789)	1.0% (17/1789)	1.5% (27/1789)	0.3% (6/1789)	0.7% (12/1789)

Performance Characteristics

Clinical Performance

Performance characteristics of the Click Test were established in two multi-center studies conducted at 14 clinical sites representative of CLIA waived testing facilities. The sites were geographically distributed across the United States and included an OB/GYN physician's office, sexual health clinics, primary care clinics, a public health clinic, a university student health clinic, an HIV/AIDS clinic, and STD clinics. A total of 32 untrained operators, representative of CLIA waived users, participated in the study.

The study subjects were prospectively enrolled females, 14 years of age and older, who self-collected vaginal swab specimens using the Visby Vaginal Specimen Collection Kit. The average age among study participants was 34 years, with a range between 14 to 80 years of age.

The table below shows the prevalence of each pathogen at each study site based on the comparator results.

Table 2. Pathogen Prevalence by Site and Overall (based on comparator results)

Site	% (# positive / # tested)		
	CT	NG	TV
1	1.6% (3/185)	0.5% (1/184)	0.0% (0/184)
2	3.9% (9/233)	1.3% (3/236)	18.9% (42/222)
3	1.5% (1/66)	1.5% (1/67)	4.6% (3/65)
4	27.2% (72/265)	9.3% (25/269)	9.8% (26/266)
5	6.6% (4/61)	4.9% (3/61)	13.3% (8/60)
6	8.6% (23/268)	0.7% (2/269)	0.7% (2/269)
7	3.7% (12/326)	0.9% (3/330)	11.2% (37/330)
8	0.0% (0/15)	6.7% (1/15)	13.3% (2/15)
9	6.8% (4/59)	1.7% (1/59)	6.8% (4/59)
10	0.0% (0/51)	0.0% (0/51)	5.9% (3/51)
11	6.4% (5/78)	2.6% (2/78)	3.9% (3/77)
12	10.5% (6/57)	1.8% (1/57)	1.8% (1/57)
13	11.1% (7/63)	1.6% (1/63)	4.8% (3/63)
14	14.9% (7/47)	2.1% (1/47)	6.4% (3/47)
Total	8.6% (153/1774)	2.5% (45/1786)	7.8% (137/1765)

The collected samples were provided to participating study operators who tested them on-site using the Click Test. The participating operators conducted the test by following the instructions in the Quick Reference Guide (QRG). The study operators had no formal training or experience with CLIA high or moderate complexity testing and did not receive any training on the use of the Visby test.

Three additional vaginal swabs were collected from each female by a licensed clinician and were sent to one central laboratory for comparator testing with three FDA cleared nucleic acid amplification tests (NAATs) detecting CT, NG and TV.

A total of 1899 subjects were initially enrolled, of which 1881 met the study inclusion criteria. Of those, 1789 females (929 symptomatic and 860 asymptomatic) were included in the performance evaluation. Study samples were excluded from the data analysis due to lack of a valid Visby test result (n=28) or for protocol deviations

(n=64), (e.g., failure to follow the study protocol, improper execution of the Visby test). Samples were also excluded from the data analysis due to lack of a valid comparator test result (CT=15, NG=3 and TV=24). Among the 1817 tests performed on the Visby Test, 119 had an invalid result on the first test, for an overall invalid rate of 6.55% (119/1817), with 95% CI (5.5%-7.8%).

The Click Test results for CT and NG were compared to a composite comparator result (CCR) comprised of results of three FDA-cleared NAATs testing clinician collected vaginal swabs. A positive (infected) comparator result for CT and NG was determined when at least two of the three comparator assays were positive. The performance estimates for the Visby Test for the detection of CT and NG were calculated as positive percent agreement (PPA) and negative percent agreement (NPA) with the composite comparator result.

The following two tables summarize the clinical performance of the Click Test for CT and NG when compared to the CCR.

Table 3. Clinical Performance of the Click Test for CT vs. Composite Comparator Results, by Symptom Status

Symptom Status	N	TP	FP	TN	FN	Prevalence%	PPA (95% CI)	NPA (95% CI)
Symptomatic	918	95	26	795	2	10.6%	97.9% (92.8-99.4%)	96.8% (95.4-97.8%)
Asymptomatic	856	54	10	790	2	6.5%	96.4% (87.9-99.0%)	98.8% (97.7-99.3%)
Overall	1774	149	36	1585	4	8.6%	97.4% (93.5-99.0%)	97.8% (96.9-98.4%)

PPA=Positive Percent agreement with CCR; NPA=Negative Percent Agreement with CCR;
TP=true positive; FP=false positive; TN=true negative; FN=false negative

Table 4. Clinical Performance of the Click Test for NG vs. Composite Comparator Results, by Symptom Status

Symptom Status	N	TP	FP	TN	FN	Prevalence %	PPA (95% CI)	NPA (95% CI)
Symptomatic	929	25	8	896	0	2.7%	100.0% (86.7-100.0%)	99.1% (98.3-99.6%)
Asymptomatic	857	19	8	829	1	2.3%	95.0% (76.4-99.1%)	99.0% (98.1-99.5%)
Overall	1786	44	16	1725	1	2.5%	97.8% (88.4-99.6%)	99.1% (98.5-99.4%)

PPA=Positive Percent agreement with CCR; NPA=Negative Percent Agreement with CCR;
TP=true positive; FP=false positive; TN=true negative; FN=false negative

The clinical performance of the Visby Test for detection of *T. vaginalis* was compared to a patient infected status (PIS) determined by testing clinician collected vaginal swabs with three FDA cleared NAATs for TV. The patient was considered infected if at least two of the three comparator assays were positive for TV. The performance estimates for the Visby Test for the detection of TV were calculated as percent sensitivity and percent specificity when compared with the PIS.

The following table summarizes the clinical performance of the Click Test for *T. vaginalis* when compared to the PIS.

Table 5. Clinical Performance of the Click Test for TV vs. PIS, by Symptom Status

Symptom Status	N	TP	FP	TN	FN	Prevalence%	Sensitivity % (95% CI)	Specificity % (95% CI)
Symptomatic	916	83	35	797	1	9.2%	98.8% (93.6%-99.8%)	95.8% (94.2%-97.0%)
Asymptomatic	849	53	18	778	0	6.2%	100% (93.2%-100.0%)	97.7% (96.5%-98.6%)
Overall	1765	136	53	1575	1	7.8%	99.3% (96.0%-99.9%)	96.7% (95.8%-97.5%)

TP=true positive; FP=false positive; TN=true negative; FN=false negative

The following table shows the comparator results that determined the infected / not infected composite comparator result for CT.

Table 6. Patient CT status by CCR

Patient Infected Status – CT	NAAT 1	NAAT 2	NAAT 3	Visby	Symptom Status	
					Symptomatic	Asymptomatic
Infected	+	+	+	+	49	23
Infected	+	+	+	-	1	0
Infected	+	+	-	+	1	1
Infected	+	+	-	-	0	1
Infected	+	+	NA[a]	+	40	29
Infected	+	+	NA[a]	-	0	1
Infected	+	-	+	+	3	0
Infected	+	-	+	-	1	0
Infected	-	+	+	+	2	1
Non-infected	+	-	-	+	1	0
Non-infected	NA[b]	-	-	-	2	1
Non-infected	-	+	-	-	4	5
Non-infected	-	NA[b]	-	-	1	0
Non-infected	-	-	+	-	0	3
Non-infected	-	-	NA[b]	+	1	0
Non-infected	-	-	-	+	14	7
Non-infected	-	-	-	-	317	366
Non-infected	-	-	NA[a]	+	10	3
Non-infected	-	-	NA[a]	-	471	415
Total					918	856

[a] Test not done

[b] Invalid test result

The following table shows the comparator results that determined the infected / not infected composite comparator result for NG.

Table 7. Patient NG status by CCR

Patient Infected Status – NG	NAAT 1	NAAT 2	NAAT 3	Visby	Symptom Status	
					Symptomatic	Asymptomatic
Infected	+	+	+	+	15	5
Infected	+	+	NA[a]	+	8	12
Infected	+	+	NA[a]	-	0	1
Infected	+	-	+	+	1	1
Infected	-	+	+	+	1	1
Non-infected	NA[b]	-	-	-	2	1
Non-infected	+	-	-	+	1	0
Non-infected	+	-	-	-	0	1
Non-infected	-	NA[b]	-	-	1	2
Non-infected	-	NA[b]	-	-	1	0
Non-infected	-	+	-	+	0	1
Non-infected	-	+	-	-	6	4
Non-infected	-	-	NA[b]	-	1	0
Non-infected	-	-	NA[b]	-	1	0
Non-infected	-	-	-	+	6	4
Non-infected	-	-	-	-	372	391
Non-infected	-	-	NA[a]	+	1	3
Non-infected	-	-	NA[a]	-	512	430
Total					929	857

[a] Test not done
[b] Invalid test result

The following table shows the patient infected status for TV based on testing vaginal swabs.

Table 8. Patient Infected Status – TV

Patient Infected Status – TV	NAAT 1	NAAT 2	NAAT 3	Visby	Symptom Status	
					Symptomatic	Asymptomatic
Infected	+	+	+	+	26	19
Infected	+	+	NA[a]	+	53	31
Infected	NA[b]	+	+	+	0	1
Infected	+	+	NA[a]	-	1	0
Infected	+	-	+	+	4	2
Non-infected	NA[b]	-	-	+	0	1
Non-infected	NA[b]	-	-	-	1	1
Non-infected	+	-	-	+	0	3
Non-infected	+	-	-	-	14	13
Non-infected	-	+	-	+	1	0
Non-infected	-	+	-	-	2	2
Non-infected	-	-	NA[b]	-	1	0
Non-infected	-	-	NA[b]	-	1	0
Non-infected	-	-	-	+	15	7
Non- infected	-	-	-	-	330	356
Non-infected	-	-	NA[a]	+	19	7
Non-infected	-	-	NA[a]	-	448	406
Total					916	849

[a] Test not done
[b] Invalid test result

Comparison study between Visby Test and Click Test

Three studies were conducted to evaluate the equivalence of the Visby Test with the Click Test.

Multicenter study with Untrained Operators

This study was conducted in CLIA Waived (CW) testing environments at three study sites. A subset of archived frozen self-collected vaginal swab patient samples that had been previously characterized in the clinical study for the Click Test were tested.

A total of 30 CT positive (based on comparator results from the clinic study for Click Test, and this is the same for other analytes and negative sample), 20 NG positive, 30 TV positive, and 33 negative vaginal swab specimens were selected for the study. Six (6) untrained study operators (two operators at each site) conducted the testing.

The frozen samples were de-identified, randomized, and blinded so that the study staff and test operators did not know the expected results. Operators thawed the samples and tested them on-site using the Visby Test by following the instructions in the Quick Reference Guide (QRG). The results of the Visby Test were compared to the recorded results of the Click Test. Of the 102 samples included in the study, two (2.4%) had an initial invalid test result. One sample was excluded from the data analysis

because it was invalid upon retesting. There were no additional exclusions. Table 9 summarize the concordance between the Click Test results and the Visby Test results.

Table 9. Clinical Performance of the Visby Test compared to the Click test (performed by untrained operators)

Target	N	TP	FP	TN	FN	PPA (95% CI)	NPA (95% CI)
CT	101	30	0	71	0	100.0% (88.6%-100.0%)	100.0% (94.9%-100.0%)
NG	101	20	0	81	0	100.0% (83.9%-100.0%)	100.0% (95.5%-100.0%)
TV	101	30	3	68	0	100.0% (88.6%-100.0%)	95.8% (88.3%-98.6%)

PPA=Positive Percent agreement; NPA=Negative Percent Agreement.
TP=true positive; FP=false positive; TN=true negative; FN=false negative.

Single Center study with trained operators

A second clinical comparison study was performed to include all frozen specimens with sufficient volume from the original clinical study of the Click Test, with a focus on testing samples representing a natural distribution of the pathogen loads among individuals, especially these with low target levels. To obtain a sufficient number of positive specimens (by comparator assays) for each analyte, these specimens were further supplemented with archived and banked frozen self-collected samples from two other previous Visby Medical specimen collections. These specimens were tested in house by seven trained operators over seven days and under variable lighting conditions. The specimens were randomized and blinded so that the test operators did not know the expected results.

A total of 359 de-identified, frozen, self-collected vaginal swab specimens were included in this study. Operators thawed the samples and tested them with both the Visby Test and the Click Test if sufficient sample volume was present. Specimens without sufficient volume to run on both devices were tested only on the Visby Test, and the original Click Test results were used for the comparison.

Of the 359 specimens included in the study, one specimen did not have sufficient volume to run on any test and thus was excluded from the study. Of the remaining 358 specimens tested on the Visby Test, 11 (3.1%) had an initial invalid test result. As a final result, seven specimens were excluded from performance calculation due to insufficient sample volume for retest or failure to obtain a valid retest result.

Therefore, a total of 351 specimens were included in the performance table. Table 10 summarizes the concordance between the Visby Test results and the Click Test results.

Table 10. Clinical Performance of the Visby Test compared to the Click Test (testing by trained operators)

Target	N	TP	FP	TN	FN	PPA (95% CI)	NPA (95% CI)
CT	351	116	3	232	0	100.0% (96.8%-100.0%)	98.7% (96.3%-99.6%)
NG	351	34	0	317	0	100.0% (89.9%-100.0%)	100.0% (98.8%-100.0%)
TV	351	100	4	240	7[a]	93.5% (87.1%-96.8%)	98.4% (95.9%-99.4%)

a. TV PIS (based on the original clinical study for Click Test) for all seven specimens were negative.

Testing spiked samples at low organism concentrations

To further evaluate the performance of the Visby Test as compared to Click Test at low levels of the target organisms, samples spiked at 1.5x LoD, 2x LoD or 3x LoD for each target in individual negative clinical matrix were prepared and tested. Twenty (20) CT positive, 20 NG positive, 20 TV positive and 20 negative samples were tested on both the Visby Test and Click Test by four trained operators at a single site. Testing was performed over two days under various lighting conditions in a randomized and blinded fashion. All 80 contrived samples yielded valid results (0% initial invalid) and are included in the final data analysis.

Table 11 summarizes the concordance of the results between the Visby Test and the Click Test.

Table 11. Performance of the Visby Test compared to the Click Test (spiked samples tested by trained operators)

Organism	Concentration	Correct Results / Total Tested	
		Click Test	Visby Test
CT	1.5x LoD	6/6	5/6
	2x LoD	10/10	9/10
	3x LoD	4/4	4/4
NG	1.5x LoD	5/6	5/6
	2x LoD	9/10	10/10
	3x LoD	4/4	4/4
TV	1.5x LoD	6/6	6/6
	2x LoD	10/10	10/10
	3x LoD	4/4	4/4
Negative	N/A	20/20	19/20*

* One device was unexpectedly positive for TV

Analytical Performance

Most analytical studies described below were performed with the Click Test and are representative and applicable to the Visby test. Two studies, including the "Comparison of Limit of Detection between the Visby Test and Click Test" and the "Reproducibility for the Visby Test" were performed on the Visby Test.

Limit of Detection

Limit of Detection (LoD) of the Click Test was determined for CT in elementary bodies per mL (EB/mL), NG in colony forming units per mL (CFU/mL), and TV in trophozoites per mL (troph/mL), from two distinct strains or serovars, seeded into clinical negative vaginal sample matrix. LoD is defined as the lowest concentration per sample that can be reproducibly distinguished from negative samples (the lowest concentration at which 95% samples are determined to be positive).

Each organism was individually seeded into negative vaginal swab sample matrix and tested in a range-finding study at five different concentrations in replicates of 20 per concentration. A single lot of negative sample matrix and a single lot of Click Tests were used throughout this study.

The LoD values for each strain were estimated by probit analysis of the results from the range-finding study. The calculated LoDs were verified by testing 20 replicates at the estimated LoD concentration and demonstrating that at least 19 out of 20 replicates were positive.

The LoD for each organism is shown in the table below.

Table 12. LoD of CT Serovars, NG strains, and TV strains in Clinical Negative Vaginal Sample Matrix

Organism	LoD
CT Serovar H (VR-879)	16.0 EB/mL
CT Serovar D (VR-885)	5.9 EB/mL
NG (ATCC 19424)	5.7 CFU/mL
NG (ATCC 49226)	6.2 CFU/mL
TV (ATCC 30001) (metronidazole susceptible)	1.2 troph/mL
TV (ATCC 30238) (metronidazole resistant)	0.24 troph/mL

Comparison of Limit of Detection between the Visby Test and Click Test

The purpose of this study is to demonstrate that the LoD for the Visby Test is equivalent to the Click Test. The LoD values were determined to be equivalent if the lowest concentrations of organism with at least 19/20 detection rate from the two devices were within 3-fold of each other. Negative pooled clinical vaginal sample in Visby Collection Media was spiked at the LoD that was established with the Click Test and tested with 20 Visby Tests and 20 Click Tests. If at least 19/20 devices returned a positive test result, the organism was diluted 3-fold and the testing was repeated until the detection rate was <19/20 for both the Click and Visby test. The results in Table 13 confirm that the LoD between the Click Test and Visby Test are comparable.

Table 13. LoD comparison between the Visby Test and the Click Test

Organism	LoD Multiple	Target Concentration	Detection Rate (Positive Valid Devices / Total Valid Devices Tested)	
			Click Test	Visby Test
CT Serovar H (VR-879)	1x	16.00 EB/mL	20/20	20/20
	1/3x	5.33 EB/mL	13/20	18/20
CT Serovar D (VR-885)	1x	5.90 EB/mL	20/20	20/20
	1/3x	1.97 EB/mL	11/20	19/20
	1/9x	0.66 EB/mL	7/20	10/20
NG (ATCC 49226)	1x	6.20 cfu/mL	19/20	20/20
	1/3x	2.06 cfu/mL	10/20	19/20
	1/9x	0.68 cfu/mL	5/20	9/20
NG (ATCC 19424)	3x	17.1 cfu/mL	20/20	20/20
	1x	5.7 cfu/mL	17/20	20/20
	1/3x	1.9 cfu/mL	17/20	19/20
	1/9x	0.63 cfu/mL	8/20	9/20
TV (ATCC 30001)[a]	1x	1.2 troph/ mL	19/20	20/20
	1/3x	0.4 troph/mL	20/20	19/20
	1/9x	0.13 troph/mL	16/20	16/20
TV (ATCC 30238)[b]	1x	0.24 troph/mL	20/20	20/20
	1/3x	0.08 troph/mL	17/20	17/20

a. Metronidazole susceptible
b. Metronidazole resistant

Inclusivity

The ability of the Click Test to detect 14 serovars of CT, 30 strains of NG, and 15 strains of TV at or near the LoD was evaluated. Each organism was individually seeded into negative clinical vaginal swab matrix at or near 2x LoD and tested in 3 replicates. If any strain did not result in 3/3 detection, then the next lower dilution was tested until detected in 3/3. All 14 CT serovars, 30 NG strains, and 15 TV strains were successfully detected at the following concentrations:

Table 14. CT Organisms Tested in Inclusivity Study

ATCC Number	Serovar	CT Concentration Tested
VR-346	F	32 EB/mL
VR-347	Ba	32 EB/mL
VR-348B	E	32 EB/mL
VR-571B	A	32 EB/mL
VR-573	B	32 EB/mL
VR-878	G	32 EB/mL
VR-880	I	128 EB/mL
VR-886	J	32 EB/mL
VR-887	K	32 EB/mL
VR-901B	LGV I	32 EB/mL
VR-902B	LGV II	64 EB/mL
VR-903	LGV III	32 EB/mL
VR-1500	C	32 EB/mL
nvCT, Swedish variant	E	32 EB/mL

Table 15. NG Organisms Tested in Inclusivity Study

ATCC Number	NG Concentration Tested	ATCC Number	NG Concentration Tested
BAA-1833	12.4 cfu/mL	27632	12.4 cfu/mL
BAA-1839	12.4 cfu/mL	27633	12.4 cfu/mL
BAA-1847	24.8 cfu/mL	31148	12.4 cfu/mL
9826	12.4 cfu/mL	31149	12.4 cfu/mL
9827	12.4 cfu/mL	31151	12.4 cfu/mL
9830	12.4 cfu/mL	31356	12.4 cfu/mL
10874	12.4 cfu/mL	31397	12.4 cfu/mL
11688	12.4 cfu/mL	31398	12.4 cfu/mL
11689	12.4 cfu/mL	31401	12.4 cfu/mL
19088	12.4 cfu/mL	31402	12.4 cfu/mL
23050	12.4 cfu/mL	31403	12.4 cfu/mL
23051	12.4 cfu/mL	31406	12.4 cfu/mL
27628	12.4 cfu/mL	35541	12.4 cfu/mL
27629	12.4 cfu/mL	43069	12.4 cfu/mL
27631	12.4 cfu/mL	49981	12.4 cfu/mL

Table 16. TV Organisms Tested in Inclusivity Study

ATCC Number	TV Concentration Tested
PRA-95	2.4 troph/mL
PRA-98	2.4 troph/mL
30184	2.4 troph/mL
30187	2.4 troph/mL
30188	2.4 troph/mL
30236	2.4 troph/mL
30240	2.4 troph/mL
30245	2.4 troph/mL
50138	2.4 troph/mL
50139	2.4 troph/mL
50141	2.4 troph/mL
50143	2.4 troph/mL
50147	2.4 troph/mL
50167	2.4 troph/mL
50183	2.4 troph/mL

Cross Reactivity and Microbial Interference

The cross reactivity of the Click Test was evaluated by testing 144 different microorganisms in replicates of 3 in negative vaginal swab matrix. Quantified stocks of whole microorganisms were spiked into the matrix and tested at high concentrations (>10^6 genomic copies/mL for bacteria and >10^5 genomic copies/mL for viruses). No cross reactivity was observed with any of the organisms tested.

Microbial Interference, when testing with the Click Test, was evaluated by testing the same 144 microorganisms in the presence of low concentrations of the target organisms (3X LoD for CT, NG and TV). No microbial interference was observed with any of the organisms tested.

Additionally, three organisms could not be obtained for direct testing (Bacterial Vaginosis Associated Bacteria 2 (BVAB-2), *Megasphaera* type 1, and *Dientamoeba fragilis*). The sequences of these organisms were analyzed against the Click Test primer and amplicon sequences using basic local alignment search tool (BLAST). No match was found for any of the 3 organisms.

Note: The organisms evaluated in the two studies are listed in the table on the following pages.

Table 17. Microorganisms Evaluated for Cross-Reactivity and Microbial Interference

Microorganism (ATCC Number)	Microorganism (ATCC Number)
Achromobacter xerosis (14780)	Eikenella corrodens (23834)
Acinetobacter calcoaceticus (23055)	Elizabethkingia meningoseptica (13253)
Acinetobacter lwoffii (15309)	Entamoeba histolytica (30458)
Actinomyces israelii (12102)	Enterococcus faecalis (29212)
Aerococcus viridans (700406)	Enterococcus faecium (19434)
Aeromonas hydrophila (35654)	Enterobacter cloacae (13047)
Alcaligenes faecalis (8750)	Enterococcus raffinosus (avium) (49464)
Arcanobacterium pyogenes (49698)	Erysipelothrix rhusiopathiae (19414)
Atopobium vaginae (BAA-55)	Escherichia coli (700928D-5)
Bacteroides fragilis (25285)	Fusobacterium nucleatum (25586)
Bacteroides ureolyticus (33387)	Gardnerella vaginalis (801894)
Bergeriella denitrificans (14686)	Gemella haemolysans (10379)
Bifidobacterium adolescentis (15703)	Giardia intestinalis (50581)
Bifidobacterium breve (15700)	Haemophilus ducreyi (33940)
Bifidobacterium longum (15697)	Haemophilus influenzae (49247)
Blastocystis hominis (50629)	Herpes simplex virus I (VR-539)
Brevibacterium linens (21330)	Herpes simplex virus II (VR-540)
BV associated bacteria (BVAB-2, N/A)*	HIV-1 (synthetic RNA) (VR-3245SD)
Campylobacter jejuni (33291)	Human papilloma virus 16 (synthetic DNA) (VR-3240SD)
Candida albicans (801504, Zeptometrix)	
Candida glabrata (90030)	Human papilloma virus 16 E6/E7 (Transforme cells) (CRL-2616)
Candida parapsilosis (22019)	
Candida tropicalis (750)	Kingella dentrificans (33394)
Chlamydophila pneumoniae (53592)	Kingella kingae (23330)
Chlamydophila psittaci (MBC013-R, Vircell)	Klebsiella aerogenes (13048)
Chlamydia trachomatis LGVII (VR-902B)**	Klebsiella oxytoca (49131)
Chromobacterium violaceum (12472)	Klebsiella pneumoniae (801506, Zeptometrix)
Citrobacter freundii (8090)	Lactobacillus acidophilus (4356)
Clostridium difficile (9689)	Lactobacillus brevis (14869)
Clostridium perfringens (13124)	Lactobacillus crispatus (33820)
Corynebacterium genitalium (33034)	Lactobacillus jensenii (25258)
Corynebacterium xerosis (373)	Lactobacillus vaginalis (49540)
Cryptococcus neoformans (66031)	Lactococcus lactis (19435)
Cryptosporidium parvum (PRA-67DQ)	Legionella pneumophila (33152, 33153)
Cutibacterium acnes (6919)	Listeria monocytogenes (19115)
Deinococcus radiodurans (13939)	Megashaera type 1 (N/A)*
Derxia gummosa (15994)	Micrococcus luteus (4698)
Dientamoeba fragilis (N/A)*	Mobiluncus curtisii (35241)

Microorganism (ATCC Number)
Mobiluncus mulieris (35243)
Moraxella lacunata (17967)
Moraxella osloensis (19976)
Moraxella (Branhamella) catarrhalis (25240)
Morganella morganii (25830)
Mycobacterium smegmatis (14468)
Mycoplasma genitalium (49123)
Mycoplasma hominis (23114)
Neisseria elongata (25295, 29315, 49378)
Neisseria cinerea (14685)
Neisseria subflava (14221)
Neisseria flavescens (13116, 13120)
Neisseria perflava (14799)
Neisseria lactamica (23970, 23971, 23972, 49142)
Neisseria meningitidis serogroup a (13077)
Neisseria meningitidis serogroup b (13090)
Neisseria meningitidis serogroup c (13102, 13105, 13112, 19577)
Neisseria meningitidis serogroup D (13113)
Neisseria meningitidis serogroup w-135 (35559)
Neisseria meningitidis serogroup y (35561)
Neisseria polysaccharea (43768)
Neisseria subflava (49275)
Neisseria mucosa (19695, 25998, 49233)
Neisseria sicca (9913, 29193, 29256)
Pantoea agglomerans (27155)
Paracoccus denitrificans (13543)
Pentatrichomonas hominis (30000)
Peptostreptococcus anaerobius (27337)
Peptostreptococcus productus (Blautia producta) (35244)

Microorganism (ATCC Number)
Plesiomonas shigelloides (51903)
Prevotella bivia (29303)
Proteus mirabilis (7002)
Proteus vulgaris (6380)
Providencia stuartii (33672)
Pseudomonas aeruginosa (801519, Zeptometrix)
Pseudomonas fluorescens (13525)
Pseudomonas putida (12633)
Rahnella aquatilis (33071)
Rhodospirillum rubrum (11170)
Saccharomyces cerevisiae (9763)
Salmonella minnesota (49284)
Salmonella typhimurium (19585)
Serratia marcescens (13880)
Staphylococcus aureus (12600)
Staphylococcus epidermidis (14990)
Staphylococcus saprophyticus (15305)
Streptococcus agalactiae (13813)
Streptococcus bovis (35034)
Streptococcus mitis (49456)
Streptococcus mutans (25175)
Streptococcus pneumoniae (6303)
Streptococcus pyogenes (19615)
Streptococcus salivarius (13419)
Streptococcus sanguinis (10556)
Streptomyces griseinus (23915)
Ureaplasma urealyticum (27618)
Trichomonas tenax (30207)
Vibrio parahaemolyticus (17802)
Yersinia enterocolitica (23715)

* These organisms were not available for direct testing and were evaluated using in-silico analysis
** This organism was correctly positive with the CT assay and negative for NG and TV

Interfering Substances

The performance of the Click Test in the presence of potentially interfering substances that may be found in a vaginal swab sample was evaluated. The potential interfering substances were diluted in the negative swab matrix and tested in the presence of low concentrations (3x LoD) of CT, NG, and TV organisms. The testing was also performed with negative clinical swab matrix samples. All samples were tested in triplicate.

The following substances were tested and found not to interfere with the assay up to the concentrations shown below.

Table 18. Potentially Interfering Substances

Substances	Concentration	Substances	Concentration
Abreva Cold Sore Cream	0.25% w/v	Replens Long Lasting Vaginal Moisturizer[c]	2.50% w/v
Beta Estradiol	0.07 mg/mL		
Biotin	3.5 µg/mL	Seminal fluid	5.00% v/v
Dove 0% alcohol anti-perspirant spray[a]	0.19% w/v	Summer's Eve Povidone- Iodine Medicated Douche	0.25% w/v
KY Jelly personal lubricant	0.25% w/v	Summer's Eve, Cleansing Wash	0.40% w/v
Leukocytes	$1x10^6$ cells/mL		
Menstrual Blood	10.0% v/v	Vaginal anti-fungal	0.25% w/v
Monistat 1	0.25% w/v	7-day Vaginal cream	0.25% w/v
Mucin (bovine)	0.80% w/v	Vagisil Moisturizer	0.25% w/v
Preparation H Hemorroidal Ointments	0.25% w/v	Vagisil Regular Strength Anti-Itch Creme	0.25% w/v
Progesterone	0.07 mg/mL	VCF Vaginal Contraceptive Gel	0.25% w/v
RepHresh Odor Eliminating pH Balancing Gel[b]	1.25% w/v	Yeast Gard Douche Advanced	0.25% w/v

[a]Dove 0% alcohol anti-perspirant spray may cause false positive results for CT, NG, and/or TV when present at a concentration greater than 0.19% (w/v)

[b] RepHresh Odor Eliminating pH Balancing Gel may cause false negative results for CT and/or NG when present at a concentration greater than 1.25% (w/v)

[c] Replens Long Lasting Vaginal Moisturizer may cause invalid results when present at a concentration greater than 2.50% (w/v)

Competitive Interference

The Click Test was evaluated for performance in the case of a mixed infection (presence of multiple target organisms). Each of the target organisms (CT, NG, and TV) were seeded into clinical negative vaginal sample matrix at varying concentrations and then tested in triplicate. Low concentrations were prepared at 3x LoD for the respective organisms and high concentrations were prepared at 1 x 10^6 units/mL. No competitive interference was observed at the levels tested for any of the three target organisms.

Table 19. Competitive Interference for each Target Organisms

Organism and Concentration			CT (# Positive / # Tested)	NG (# Positive / # Tested)	TV (# Positive / # Tested)
CT	NG	TV			
Low	Low	Low	3/3	3/3	3/3
Low	High	High	3/3	3/3	3/3
Low	High	Neg	3/3	3/3	0/3
Low	Neg	High	3/3	0/3	3/3
High	Low	High	3/3	3/3	3/3
High	Low	Neg	3/3	3/3	0/3
Neg	Low	High	0/3	3/3	3/3
High	High	Low	3/3	3/3	3/3
Neg	High	Low	0/3	3/3	3/3
High	Neg	Low	3/3	0/3	3/3
High	Neg	Neg	3/3	0/3	0/3
Neg	High	Neg	0/3	3/3	0/3
Neg	Neg	High	0/3	0/3	3/3

Reproducibility

A reproducibility study was performed to evaluate the reproducibility of the Click Test when used by untrained users in CLIA waived settings. The operators performing the testing were non-laboratorians representing healthcare professionals that may be encountered at such sites. The study evaluated four (4) panel members that were prepared using cultured organisms in negative pooled clinical vaginal swab matrix (previously determined to be negative for CT, NG, TV). The study was performed with negative (unspiked) and positive samples.

A total of six (6) study operators (2 operators at each site) tested the panel three (3) times each testing day, over six (6) non-consecutive days. Three reagent lots were used in the study. A summary of the results (count correct / total count) and % agreement with expected results for each analysis is presented in the table below. The Click Test demonstrated robust reproducibility with no significant effect observed for the components of variation evaluated (sites, days, operators, lots).

Table 20. Summary of Reproducibility Results with Click Test

	Site 1	Site 2	Site 3	Overall Agreement	
Panel Member	**% Agreement (count)**	**% Agreement (count)**	**% Agreement (count)**	**% Agreement (count)**	**95% CI**
CT Positive (49.72 EB/mL)	100% (35/35) [a]	100% (36/36) [b]	100% (36/36) [c]	100% (107/107)	92.1%-99.0%
NG Positive (22.68 cfu/mL)	97.1% (34/35) [a]	94.3% (33/35) [a]	100% (36/36)	97.2% (103/106)	92.0%-99.0%
TV Positive (21.6 troph/mL)	100% (36/36)	100% (35/35) [a]	100% (35/35) [a]	100% (106/106)	96.5%-100%
Negative	97.2% (35/36) [d]	100% (36/36)	97.2% (35/36) [e]	98.1% (106/108)	93.5%-99.5%

[a] One sample had invalid results and was omitted from the analysis.
[b] One sample was positive result for CT, but unexpectedly positive for NG and TV
[c] Two samples were Ct positive, but unexpected positive for TV
[d] One sample was unexpectedly positive for TV
[e] One sample was unexpectedly positive for NG

An additional study using low positive (spiked at ~1X LoD) and negative (unspiked) samples was performed to evaluate the repeatability of test results near the assay LoD when tested by untrained operators in a CLIA waived setting. A total of six (6) operators (2 operators at each site) tested the panel twice a day for 5 days, for a total of 60 data points per panel member. The composition of the panel members along with summary of results (count correct / total count) and percent agreement with the expected result for each panel member is presented in the table below. The study demonstrated that untrained users can perform the test and interpret the results accurately when testing samples with organism concentrations near the LoD.

Table 21. Summary of Study with Samples Near Assay LoD

	Site 1	Site 2	Site 3	Overall Agreement	
Panel Member	**% Agreement (count)**	**% Agreement (count)**	**% Agreement (count)**	**% Agreement (count)**	**95% CI**
CT Low Positive (16.0 EB/mL)	100% (20/20)	100% (20/20)	100% (20/20)	100% (60/60)	94.0%-100%
NG Low Positive (6.2 cfu/mL)	95.0% (19/20)	95.0% (19/20)	100% (20/20)	96.7% (58/60)	88.6%-99.1%
TV Low Positive (1.2 troph/mL)	100% (20/20)	95.0% (19/20)	95.0% (19/20)	96.7% (58/60)	88.6%-99.1%
Negative	100% (18/18)*	100% (20/20)	100% (20/20)	100% (58/58)	93.8%-100%

*Two samples had invalid results and were omitted from the analysis.

Reproducibility for the Visby Test

A reproducibility study was conducted at three (3) study sites with CLIA Waived settings by untrained operators. The operators performing the testing were non-laboratorians representing healthcare professionals that may be encountered at such sites. The study evaluated seven (7) panel members that were prepared using cultured organisms in negative pooled clinical vaginal swab matrix (previously determined to be negative for CT, NG, TV). The study was performed with negative (unspiked), low positive (1XLoD), and moderate positive (3-5 X LoD) samples.

A total of six (6) study operators (two operators at each site) tested the panel three (3) times each testing day, over six (6) non-consecutive days. Three reagent lots were used in the study. A summary of the results (count correct / total count) and % agreement with expected results for each panel member by site and overall is presented in the table below. The Visby Test demonstrated that untrained users can perform testing accurately and reproducibly.

Table 22. Summary of Reproducibility Study Results for the Visby Test

Panel Member	Site 1	Site 2	Site 3	Overall Agreement	
	% Agreement (count)	% Agreement (count)	% Agreement (count)	% Agreement (count)	95% CI
CT Moderate Positive (64.0 EB/mL)	100.0% (36/36)	100.0% (36/36)	100.0% (36/36)	100.0% (108/108)	96.6%-100.0%
CT Low Positive (16.0 EB/mL)	97.2% (35/36)	100.0% (36/36) [a]	100.0% (36/36)	99.1% (107/108)	94.9%-99.8%
NG Moderate Positive (24.8 cfu/mL)	100.0% (36/36)	100.0% (36/36)	97.2% (35/36)	99.1% (107/108)	94.9%-99.8%
NG Low Positive (6.2 cfu/mL)	100.0% (36/36)	100.0% (36/36)	100.0% (36/36)	100.0% (108/108)	96.6%-100.0%
TV Moderate Positive (4.8 troph/mL)	100.0% (36/36)	100.0% (36/36)	100.0% (36/36)	100.0% (108/108)	96.6%-100.0%
TV Low Positive (1.2 troph/mL)	97.2% (35/36)	97.2% (35/36) [b]	94.4% (34/36)	96.3% (104/108)	90.9%-98.6%
Negative	97.2% (35/36) [c]	100.0% (36/36)	97.2% (35/36) [c]	98.1% (106/108)	93.5%-99.5%

[a] One CT Low Positive sample was unexpectedly positive for TV

[b] One TV Low Positive sample was unexpected positive for CT

[c] One Negative sample was unexpectedly positive for TV

EMC Compliance

The Visby Test was tested and found compliant in full to all requirements of IEC 60601-1-2: 2014 +A1: 2020. The compliance for each emissions and immunity standard or test specified by this collateral standard is provided in the tables below:

Table 23: Electromagnetic Emissions

Emissions Test	Compliance
Conducted Emissions Mains Terminal Measurements	IEC 60601-1-2 CISPR 11 Group 1, Class B
Radiated Emissions Measurements	IEC 60601-1-2 CISPR 11 Group 1, Class B
Harmonic Current Emissions	IEC 61000-3-2
Voltage Fluctuations / Flicker Emissions	IEC 61000-3-3

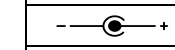
Table 24: Electromagnetic Immunity

Immunity Test / Basic EMC Standard or Test Method	IEC 60601-1-2: 2014 Test Levels
Electrostatic Discharge (ESD) Immunity IEC 61000-4-2	Contact: +/- 8 kV Air: +/- 2 kV; +/- 4 kV; +/- 8 kV; +/- 15 kV
Radiated Immunity IEC 61000-4-3	10 V/m over the frequency ranges of 80 MHz to 2,700 MHz, modulated with 1 kHz at 80% amplitude modulation RF wireless communications equipment: 385 MHz to 5785 MHz
Electrical Fast Transient/Burst Immunity IEC 61000-4-4	Power Supply Lines: ±2 kV with a 100 kHz repetition rate
Surge Immunity IEC 61000-4-5	Line to Line: ±0.5 kV, ±1 kV
Conducted Immunity IEC 61000-4-6	3 Vrms: 1 kHz at 80% amplitude modulation injection over the frequency range 150 kHz to 80 MHz 6 Vrms: 1 kHz at 80% amplitude modulation injection over the frequency range 150 kHz to 80 MHz
Power Frequency Magnetic Field Immunity (60 Hz) IEC 61000-4-8	30 A/m at 60 Hz
Voltage Dips, Short Interruptions, and Voltage Variations Immunity IEC 61000-4-11	Dips: 0% of the rated voltage (100V and 240V) for 0.5 cycle and 1 cycle 70% of the rated voltage (100V and 240V) for 30 cycles at 60 Hz Interruptions: 0 % of the rated voltage (100V and 240V) for 300 cycles at 60 Hz

References

1. Gonorrhea detailed factsheet [Internet]. Centers for Disease Control and Prevention; accessed updated 2021 July 22. Available from: https://www.cdc.gov/std/gonorrhea/stdfactgonorrhea-detailed.htm.
2. Chlamydia detailed factsheet [Internet]. Centers for Disease Control and Prevention; updated 2021 July 22. Available from: https://www.cdc.gov/std/chlamydia/stdfactchlamydia-detailed.htm.
3. Trichomoniasis factsheet [Internet]. Centers for Disease Control and Prevention; updated 2021 July 22. Available from: https://www.cdc.gov/std/trichomonas/stdfacttrichomoniasis. htm.
4. Morré, S. A., Rozendaal, L., van Valkengoed, I. G., Boeke, A. J., van Voorst Vader, P. C., Schirm, J., de Blok, S., van Den Hoek, J. A., van Doornum, G. J., Meijer, C. J., & van Den Brule, A. J. (2000). Urogenital Chlamydia trachomatis serovars in men and women with a symptomatic or asymptomatic infection: an association with clinical manifestations? JCM, 38(6), 2292–2296. https://doi.org/10.1128/JCM.38.6.2292-2296.2000
5. Centers for Disease Control and Prevention. Biosafety in microbiological and biomedical laboratories. 6th Edition.
6. CLSI Publication M29. Protection of laboratory workers from occupationally acquired infections; Approved Guideline. 4th Edition.

Index of Symbols

Symbol	Meaning	ISO 15223-1 Ref. Number
	Power supply (9 V, 3.5 A DC, 31.5 W)	N/A
	Catalog number	5.1.6
	Do not reuse	5.4.2
	Handle with care	5.3.1
	Batch code	5.1.5
	Caution	5.4.4
	Consult instructions for use	5.4.3
	Manufacturer	5.1.1
	Expiration date	5.1.4
	Temperature limitation	5.3.7
	Humidity limitation	5.3.8
	Biological risk	5.4.1
	In vitro diagnostic medical device	5.5.1
	Do no use if package is damaged	5.2.8
	Nemko 61010	N/A
	Prescription Use Only	N/A
	Positive / negative controls	5.5.4 / 5.5.3
	Atmospheric Pressure limitation	5.3.9

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