Rx Only

cobas® CT/NG

Qualitative nucleic acid test for use on the cobas® 6800/8800 Systems

For in vitro diagnostic use

cobas® CT/NG	P/N: 07460066190
cobas® CT/NG Positive Control Kit	P/N: 07460082190
cobas® Buffer Negative Control Kit	P/N: 07002238190

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Intended use

cobas® CT/NG for use on the **cobas®** 6800/8800 Systems is an automated, qualitative *in vitro* nucleic acid diagnostic test, that utilizes real-time polymerase chain reaction (PCR), for the direct detection of *Chlamydia trachomatis* (CT) and/or *Neisseria gonorrhoeae* (NG) 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 Media (Roche Molecular Systems, Inc.), and cervical specimens collected in PreservCyt® Solution. This test is intended as an aid in the diagnosis of chlamydial and gonococcal disease in both symptomatic and asymptomatic individuals.

Summary and explanation of the test

Background

Infection with CT is the leading bacterial cause of sexually transmitted diseases worldwide, with approximately 89.1 million cases occurring annually.[1] *C. trachomatis* is the most frequently reported bacterial sexually transmitted disease (STD) in the United States[1,2] and prevalence is highest in persons aged ≤ 24 years.[3] In 2013, a total of 1,401,906 cases of *C. trachomatis* infection were reported to the CDC corresponding to a rate of 446.6 cases per 100,000 population.[3] CT is a gram-negative, nonmotile, obligate intracellular bacterium with a unique biphasic lifecycle.[1] CT causes a variety of infections including urethritis, cervicitis, proctitis, conjunctivitis, endometritis, and salpingitis; if left untreated, the infection may ascend to the uterus, fallopian tubes, and ovaries causing pelvic inflammatory syndrome, ectopic pregnancy, and tubal factor infertility. Reiter's syndrome (urethritis, conjunctivitis, arthritis, and mucocutaneous lesions) has also been associated with genital CT infection.[1] Many infections remain asymptomatic, and high numbers of infected patients may not seek care.[4] Patients often become re-infected if their sexual partners are not treated. Infants born to infected mothers can develop conjunctivitis, pharyngitis, and pneumonia. The predominant symptoms in men and women are increased discharge and dysuria; women may also present with irregular uterine bleeding.[1]

The diagnosis of *C. trachomatis* urogenital infection in women is made by testing first-catch urine or collecting swab specimens from the endocervix or vagina. Diagnosis of *C. trachomatis* urethral infection in men can be made by testing a urethral swab or first-catch urine specimen. Nucleic acid amplification tests (NAATs) are the most sensitive tests for these specimens and therefore are recommended for detecting *C. trachomatis* infection.[5]

Annual screening for CT of all sexually active women aged < 25 years is recommended and screening of older women is recommended in the presence of increased risk for infection (e.g., those who have a new sex partner, more than one sex partner, a sex partner with concurrent partners, or a sex partner who has a sexually transmitted infection).[6] Chlamydia screening programs have been demonstrated to reduce the rates of PID in women.[7,8] Although the evidence to support routine screening for CT in sexually active young men is insufficient, due to the relative lack of feasibility, efficacy, and cost-effectiveness studies, the screening of sexually active young men should be considered in clinical settings with a high prevalence of chlamydia (e.g., adolescent clinics, correctional facilities, and STD clinics) or in populations with high burden of infection (e.g., men who have sex with men [MSM]).[2,6] The primary focus of chlamydia screening efforts among women should be to detect chlamydia, prevent complications, and test and treat their partners, whereas targeted chlamydia screening in men should only be considered when resources permit, prevalence is high, and such screening does not hinder chlamydia screening efforts in women.[9,10] More frequent screening for some women (e.g., adolescents) or certain men (e.g., MSM) might be indicated.[2]

Neisseria gonorrhoeae (NG) is the etiologic agent of gonorrhea and are cytochrome oxidase-positive, non-motile, non-spore

forming gram-negative diplococci. In the United States, an estimated 820,000 new *N. gonorrhoeae* infections occur each year.[11] Gonorrhea is the second most commonly reported communicable disease.[3] Clinical manifestations of NG infections are numerous.[4] In men, acute urethritis presents itself after a 1-10 day incubation period with urethral discharge and dysuria. Only a small proportion of men remain asymptomatic without signs of urethritis.[12] Acute epididymitis is the most common complication, especially in young men. In women, the primary site of infection is the endocervix. There is a high prevalence of coalescence of symptoms with CT, *Trichomonas vaginalis*, and vaginosis; many women remain asymptomatic and therefore do not seek medical care. In symptomatic women increased discharge, dysuria, and intermenstrual bleeding may be observed.[13] Pelvic inflammatory disease (PID) can occur in 10%-20% of women, combined with endometritis, salpingitis, tubo-ovarian abscess, pelvic peritonitis, and perihepatitis.[14] PID can result in tubal scarring that can lead to infertility and ectopic pregnancy. Other gonococcal infected sites in men and women are the rectum, pharynx, conjunctiva, and to a lesser degree the disease presents itself as disseminated gonococcal infection. Infants from infected mothers can develop conjunctivitis.

Annual screening for *N. gonorrhoeae* infection is recommended for all sexually active women aged < 25 years and for older women at increased risk for infection (e.g., those who have a new sex partner, more than one sex partner, a sex partner with concurrent partners, or a sex partner who has an STI).[6] Additional risk factors include inconsistent condom use among persons with multiple sex partners, previous or coexisting sexually transmitted infections, and exchanging sex for money or drugs.[2] In addition to urethral infections, the CDC also recommends the use of NAATs for routine annual screening for MSM for anorectal or oral infection.[5]

Rationale for CT/NG testing

NAATs are the recommended method for CT and NG screening.[15] For women, a vaginal swab is the recommended sample type and first catch urine is recommended for men. Alternative acceptable sample types for women include an endocervical swab when a pelvic examination is indicated or a first catch urine sample, but a urine sample may detect up to 10% fewer infections when compared with vaginal and endocervical swabs. In addition to urine for men, a urethral swab is also acceptable.

cobas® CT/NG for use on the **cobas**® 6800/8800 Systems (referred to as **cobas**® CT/NG throughout the remainder of this document) is an automated, qualitative real-time PCR test designed to detect CT and NG DNA in urogenital specimens from male and female patients and thus fulfills the medical need for a rapid, high throughput molecular screening test for use as an aid in the diagnosis of chlamydial and gonococcal disease in both symptomatic and asymptomatic individuals.

Explanation of the test

cobas® CT/NG is a qualitative test performed on the **cobas**® 6800 System and **cobas**® 8800 System. **cobas**® CT/NG enables the detection of CT/NG DNA in endocervical, vaginal, urine and cervical specimens of infected female patients and urine specimens in infected male patients. Target-specific primers and two probes are used to detect but not discriminate between the CT 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 NG DR-9 region. The DNA Internal Control, used to monitor the entire sample preparation and PCR amplification process, is introduced into each specimen during sample processing. In addition, the test utilizes a low titer positive and a negative control.

Principles of the procedure

cobas® CT/NG is based on fully automated sample preparation (nucleic acid extraction and purification) followed by PCR amplification and detection. The **cobas**® 6800/8800 Systems consist of the sample supply module, the transfer module, the

processing module, and the analytic module. Automated data management is performed by the **cobas**® 6800/8800 software which assigns test results for all tests as positive, negative or invalid. Results can be reviewed directly on the system screen, exported, or printed as a report.

Nucleic acid from patient samples and added internal control DNA (DNA-IC) molecules is simultaneously extracted. In summary, bacterial nucleic acid is released by addition of proteinase and lysis reagent to the sample. The released nucleic acid binds to the silica surface of the added magnetic glass particles. Unbound substances and impurities, such as denatured protein, cellular debris and potential PCR inhibitors are removed with subsequent wash steps and purified nucleic acid is eluted from the magnetic glass particles with elution buffer at elevated temperature. External controls (positive and negative) are processed in the same way with each **cobas**® CT/NG run.

Selective amplification of target nucleic acid from the sample is achieved by the use of target-specific forward and reverse primers which are selected from highly conserved plasmid and genomic regions of CT and NG. A region on the CT cryptic plasmid and the ompA gene (dual target) and two conserved sequences of the NG DR-9 region are amplified by **cobas**® CT/NG. Selective amplification of DNA-IC is achieved by the use of sequence-specific forward and reverse primers which are selected to have no homology with either the CT or NG target regions. A thermostable DNA polymerase enzyme is used for PCR amplification. The target and DNA-IC sequences are amplified simultaneously utilizing a universal PCR amplification profile with predefined temperature steps and number of cycles. The master mix includes deoxyuridine triphosphate (dUTP), instead of deoxythimidine triphosphate (dTTP), which is incorporated into the newly synthesized DNA (amplicon). Any contaminating amplicons from previous PCR runs are eliminated by the AmpErase enzyme, which is included in the PCR master mix, during the first thermal cycling step.[16] However, newly formed amplicons are not eliminated since the AmpErase enzyme is inactivated once exposed to temperatures above 55°C.

The **cobas**® CT/NG master mix contains two detection probes specific for the CT target sequences, two detection probes specific for the NG target sequences and one for the DNA-IC. The probes are labeled with target specific fluorescent reporter dyes allowing simultaneous detection of CT targets, NG targets and DNA-IC in three different target channels.[17,18] When not bound to the target sequence, the fluorescent signal of the intact probes is suppressed by a quencher dye. During the PCR amplification step, hybridization of the probes to the specific single-stranded DNA template results in cleavage of the probe by the 5' to 3' exonuclease activity of the DNA polymerase resulting in separation of the reporter and quencher dyes and the generation of a fluorescent signal. With each PCR cycle, increasing amounts of cleaved probes are generated and the cumulative signal of the reporter dye increases concomitantly. Real-time detection and discrimination of PCR products is accomplished by measuring the fluorescence of the released reporter dyes for the CT and NG targets and DNA-IC, respectively.

Reagents and materials

Refer to the **Reagents and materials** section and **Precautions and handling requirements** section for the hazard information for the product.

cobas® CT/NG reagents and controls

All unopened reagents and controls shall be stored as recommended in Table 1 to Table 4.

Table 1 cobas® CT/NG

cobas® CT/NG
Store at 2-8°C
480 test cassette (P/N 07460066190)

Kit components	Reagent ingredients	Quantity per kit 480 tests
Proteinase Solution (PASE)	Tris buffer, < 0.05% EDTA, Calcium chloride, Calcium acetate, 8% Proteinase EUH210: Safety data sheet available on request. EUH208: Contains Subtilisin, 9014-01-1. May produce an allergic reaction.	38 mL
DNA Internal Control (DNA-IC)	Tris buffer, < 0.05% EDTA, < 0.001% non-CT/NG related DNA construct containing primer and probe specific sequence regions, < 0.1% Sodium azide	38 mL
Elution Buffer (EB)	Tris buffer, 0.2% Methyl-4 hydroxibenzoate	38 mL
Master Mix Reagent 1 (MMX-R1)	Manganese acetate, Potassium hydroxide, < 0.1% Sodium azide	14.5 mL
CT/NG Master Mix Reagent 2 (CT/NG MMX-R2)	Tricine buffer, Potassium acetate, EDTA, Glycerol, < 18% Dimethyl sulfoxide, < 0.12% dATP, dCTP, dGTP, dUTPs, < 0.1% Tween 20, < 0.1% Sodium azide, < 0.1% Z05 DNA polymerase, < 0.10% AmpErase (uracil-N glycosylase) enzyme (microbial), < 0.01% Internal Control forward and reverse primers, < 0.01% Upstream and downstream CT/NG primers, < 0.01% Fluorescent-labeled oligonucleotide probes specific for CT, NG and the DNA Internal Control, < 0.01% Oligonucleotide aptamer	17.5 mL

Table 2 cobas® CT/NG Positive Control Kit

cobas® CT/NG Positive Control Kit
Store at 2–8°C
(P/N 07460082190)

Kit components	Reagent ingredients	Quantity per kit
CT/NG Positive Control (CT/NG (+) C)	Tris buffer, < 0.05% Sodium azide, < 0.005% EDTA, < 0.003% Poly rA, <0.01% Non-infectious plasmid DNA (microbial) containing *C. trachomatis*, <0.01% Non-infectious plasmid DNA (microbial) containing *N. gonorrhoeae*	16 mL (16 x 1mL)

Table 3 cobas® Buffer Negative Control Kit

cobas® Buffer Negative Control Kit
Store at 2-8°C
(P/N 07002238190)

Kit components	Reagent ingredients	Quantity per kit
cobas® Buffer Negative Control (BUF (-) C)	Tris buffer, < 0.1% sodium azide, EDTA, < 0.002% Poly rA RNA (synthetic)	16 mL (16 x 1mL)

cobas omni reagents for sample preparation

Table 4 cobas omni reagents for sample preparation*

Reagents	Reagent ingredients	Quantity per kit	Safety symbol and warning**
cobas omni MGP Reagent (MGP) Store at 2–8°C (P/N 06997546190)	Magnetic glass particles, Tris buffer, 0.1% methyl-4 hydroxybenzoate, < 0.1% sodium azide	480 tests	Not applicable
cobas omni Specimen Diluent (SPEC DIL) Store at 2–8°C (P/N 06997511190)	Tris buffer, 0.1% methyl-4 hydroxybenzoate, < 0.1% sodium azide	4 x 875 mL	Not applicable
cobas omni Lysis Reagent (LYS) Store at 2–8°C (P/N 06997538190)	42.56% (w/w) guanidine thiocyanate, 5% (w/v) polydocanol, 2% (w/v) dithiothreitol, dihydro sodium citrate	4 x 875 mL	  **DANGER** H302 + H332: Harmful if swallowed or if inhaled. H314: Causes severe skin burns and eye damage. H412: Harmful to aquatic life with long lasting effects. EUH032: Contact with acids liberates very toxic gas. P261: Avoid breathing dust/ fume/ gas/ mist/ vapours/ spray. P273: Avoid release to the environment. P280: Wear protective gloves/protective clothing/eye protection/ face protection. P303 + P361 + P353: IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water. P304 + P340 + P310: IF INHALED: Remove person to fresh air and keep comfortable for breathing. Immediately call a POISON CENTER/doctor. P305 + P351 + P338 + P310: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/doctor.
cobas omni Wash Reagent (WASH) Store at 15–30°C (P/N 06997503190)	Sodium citrate dihydrate, 0.1% methyl-4 hydroxybenzoate	4.2 L	Not applicable

* These reagents are not included in the **cobas**® CT/NG kit. See listing of additional materials required (Table 7).

**Product safety labeling primarily follows EU GHS guidance

Reagent storage and handling requirements

Reagents shall be stored and handled as specified in Table 5 and Table 6.

When reagents are not loaded on the **cobas**® 6800/8800 Systems, store them at the corresponding temperature specified in Table 5.

Table 5 Reagent storage (when reagent is not on the system)

Reagent	Storage temperature
cobas® CT/NG	2–8°C
cobas® CT/NG Positive Control Kit	2–8°C
cobas® Buffer Negative Control Kit	2–8°C
cobas omni Lysis Reagent	2–8°C
cobas omni MGP Reagent	2–8°C
cobas omni Specimen Diluent	2–8°C
cobas omni Wash Reagent	15–30°C

Reagents loaded onto the **cobas**® 6800/8800 Systems are stored at appropriate temperatures and their expiration is monitored by the system. The **cobas**® 6800/8800 Systems allow reagents to be used only if all of the conditions shown in Table 6 are met. The system automatically prevents use of expired reagents. Table 6 allows the user to understand the reagent handling conditions enforced by the **cobas**® 6800/8800 Systems.

Table 6 Reagent expiry conditions enforced by the **cobas**® 6800/8800 Systems

Reagent	Open-kit stability	Number of runs for which this kit can be used	On-board stability (cumulative time on board outside refrigerator)
cobas® CT/NG	90 days from first usage	Max 20 runs	Max 20 hours
cobas® CT/NG Positive Control Kit	Not applicable	Not applicable	Max 10 hours
cobas® Buffer Negative Control Kit	Not applicable	Not applicable	Max 10 hours
cobas omni Lysis Reagent	30 days from loading*	Not applicable	Not applicable
cobas omni MGP Reagent	30 days from loading*	Not applicable	Not applicable
cobas omni Specimen Diluent	30 days from loading*	Not applicable	Not applicable
cobas omni Wash Reagent	30 days from loading*	Not applicable	Not applicable

* Time is measured from the first time that reagent is loaded onto the **cobas**® 6800/8800 Systems.

Additional materials required

Table 7 Materials and consumables for use on **cobas**® 6800/8800 Systems

Material	P/N
cobas omni Processing Plate	05534917001
cobas omni Amplification Plate	05534941001
cobas omni Pipette Tips	05534925001
cobas omni Liquid Waste Container	07094388001
cobas omni Lysis Reagent	06997538190
cobas omni MGP Reagent	06997546190
cobas omni Specimen Diluent	06997511190
cobas omni Wash Reagent	06997503190
Solid Waste Bag	07435967001
Solid Waste Container	07094361001
cobas® PCR Media Secondary Tube Kit	07958048190
cobas® PCR Media Tube Replacement Cap Kit	07958056190
Replacement Caps for PreservCyt® Vials	08037230190
cobas® PCR Media Disposable Tube Stand (Optional)	07958064190
MPA RACK 16 MM LIGHT GREEN 7001-7050[a,b]	03143449001
RD5 RACK – RD Standard rack 0001-0050 LR[a,b]	11902997001

[a] MPA 16mm and RD5 racks are required to use **cobas**® CT/NG. Contact your local Roche representative for a detailed order list for sample racks, racks for clotted tips and rack trays accepted on the instruments.

[b] MPA 16mm rack is the preferred rack. If RD5 racks are used, ensure sample tubes are filled with the recommended minimum sample input volume. Rationale: The tubes sit higher in an RD5 rack because of the rubber gasket at the bottom of each tube position. Because of this, it is possible that when using RD5 racks, the system could accept tubes that are below the minimum

Table 8 Specimen collection kits used with **cobas**® CT/NG

Collection Kit	P/N
cobas® PCR Urine Sample Kit	05170486190
cobas® PCR Media Uni Swab Sample Kit	07958030190
cobas® PCR Media Dual Swab Sample Kit	07958021190
ThinPrep Pap Test Physician's Kit (500 vials & Broom-like collection devices) ThinPrep Pap Test Physician's Kit (500 vials & Cytobrush/spatula collection devices)	Hologic: 70136-001 Hologic: 70136-002

Instrumentation and software required

The **cobas**® 6800/8800 software and **cobas**® CT/NG analysis packages (ASAPs) shall be installed on the instrument(s). The Instrument Gateway (IG) server will be provided with the system.

Table 9 Instrumentation

Equipment	P/N
cobas® 6800 System (Moveable Platform)	05524245001 and 06379672001
cobas® 6800 System (Fixed Platform)	05524245001 and 06379664001
cobas® 8800 System	05412722001
Sample Supply Module	06301037001
Instrument Gateway	06349595001

cobas® CT/NG accepts the primary tube used for all **cobas**® PCR CT/NG swab and urine specimen types. Refer to the **cobas**® 6800/8800 Systems User Guide for additional information for primary and secondary sample tubes accepted on the instruments.

Precautions and handling requirements

Warnings and precautions

As with any test procedure, good laboratory practice is essential to the proper performance of this assay. Due to the high sensitivity of this test, care should be taken to keep reagents and amplification mixtures free of contamination.

- For *in vitro* diagnostic use only.
- All patient samples should be handled as if infectious, using good laboratory procedures as outlined in Biosafety in Microbiological and Biomedical Laboratories and in the CLSI Document M29-A4.[19,20] Only personnel proficient in handling infectious materials and in the use of **cobas**® CT/NG and **cobas**® 6800/8800 Systems should perform this procedure.
- All human-sourced materials should be considered potentially infectious and should be handled with universal precautions
- Do not freeze any samples.
- Use only supplied or specified required consumables to ensure established test performance.
- Safety Data Sheets (SDS) are available on request from your local Roche representative.
- Closely follow procedures and guidelines provided to ensure that the test is performed correctly. Any deviation from the procedures and guidelines may affect established test performance.
- False positive results may occur if carryover of samples is not adequately controlled during sample handling and processing.
- **cobas**® PCR Media (from primary specimen tube) contains guanidine hydrochloride. **Do not allow direct contact between guanidine hydrochloride and sodium hypochlorite (bleach) or other highly reactive reagents such as acids or bases. These mixtures can release a noxious gas**. If liquid containing guanidine hydrochloride is spilled, clean with suitable laboratory detergent and water. If the spilled liquid contains potentially infectious agents, **FIRST** clean the affected area with laboratory detergent and water, and then with 0.5% sodium hypochlorite.

Reagent handling

- Handle all reagents, controls, and samples according to good laboratory practice in order to prevent carryover of samples, reagents, or controls.
- Before use, visually inspect each reagent cassette, diluent, lysis reagent, and wash reagent to ensure that there are no signs of leakage. If there is any evidence of leakage, do not use that material for testing.
- **cobas omni** Lysis Reagent contains guanidine thiocyanate, a potentially hazardous chemical. Avoid contact of reagents with the skin, eyes, or mucous membranes. If contact does occur, immediately wash with generous amounts of water; otherwise, burns can occur.
- Expended control kits contain pierced vials with residual reagent; special care should be taken during disposal to avoid spills and contact.
- **cobas**® CT/NG kit, **cobas**® CT/NG Positive Control kit, **cobas**® Buffer Negative Control kit, **cobas omni** MGP Reagent, and **cobas omni** Specimen Diluent contain sodium azide as a preservative. Avoid contact of reagents with the skin, eyes, or mucous membranes. If contact does occur, immediately wash with generous amounts of water; otherwise, burns can occur. If these reagents are spilled, dilute with water before wiping dry.
- **Do not allow cobas omni Lysis Reagent, which contains guanidine thiocyanate, to contact sodium hypochlorite (bleach) solution. This mixture can produce a highly toxic gas.**
- Dispose of all materials that have come in contact with samples and reagents in accordance with country, state, and local regulations.

Good laboratory practice

- Do not pipette by mouth.
- Do not eat, drink, or smoke in designated work areas.
- Wear laboratory gloves, laboratory coats, and eye protection when handling samples and reagents. Avoid contaminating gloves when handling samples and controls. Gloves must be changed between handling samples and **cobas**® CT/NG kit, **cobas**® CT/NG Positive Control kit, **cobas**® Buffer Negative Control kit**,** and **cobas omni** reagents to prevent contamination.
- Wash hands thoroughly after handling samples and reagents, and after removing the gloves.
- Thoroughly clean and disinfect all laboratory work surfaces with a freshly prepared solution of 0.5% sodium hypochlorite in distilled or deionized water (dilute household bleach 1:10). Follow by wiping the surface with 70% ethanol.
- If spills occur on the **cobas**® 6800/8800 Systems, follow the instructions in the **cobas**® 6800/8800 Systems User Guide to properly clean and decontaminate the surface of instrument(s).

Specimen collection, transport, and storage

Note: Handle all samples and controls as if they are capable of transmitting infectious agents.

Specimen collection

Endocervical swab specimens collected with the **cobas**® PCR Media Dual Swab Sample Kit, vaginal swab specimens collected with either the **cobas**® PCR Media Uni Swab Sample Kit or **cobas**® PCR Media Dual Swab Sample Kit, male and female urine collected with the **cobas**® PCR Urine Sample Kit and cervical specimens collected in PreservCyt® Solution have been validated for use with **cobas**® CT/NG (see Table 8 for a list of all collection kits). Follow the instructions for collecting swab and urine specimens in their respective collection kit IFU. Follow the manufacturer's instructions for collecting cervical specimens into PreservCyt® Solution.

Specimen transport

All specimen types listed in the "Specimen collection" section above can be transported at 2-30°C. Transportation of CT/NG specimens in **cobas**® PCR Media and PreservCyt® Solution must comply with country, federal, state and local regulations for the transport of etiologic agents.[21]

Specimen storage

Store specimens as shown in Table 10. PreservCyt® and **cobas**® PCR Media specimens should not be frozen.

Table 10 Summary of acceptable specimen storage conditions prior to testing with **cobas**® CT/NG

Specimen Type		**2–30°C**
Samples in **cobas**® PCR Media		12 months
PreservCyt®	in collection device	12 months
	or	
	PreservCyt® samples aliquoted to secondary tubes	31 days

Male and female urine specimens

- Use only the **cobas**® PCR Urine Sample Kit to collect urine specimens for **cobas**® CT/NG. **cobas**® CT/NG has not been validated for use with other urine collection devices or media types. Using **cobas**® CT/NG with other urine collection devices or other media types may lead to false negative, false positive, and/or invalid results.
- To avoid cross contamination of processed specimens, additional caps for **cobas**® PCR Media tubes in an alternate color (neutral; see **Additional materials required**) should be used to recap specimens after processing.
- Untested urine specimens must show the top of the liquid level between the two black lines on the **cobas**® PCR Media tube label window. If the liquid level is above or below these lines, the specimen has not been collected properly and cannot be used for testing.
- If additional testing is required, ensure that there is at least 1.2 mL of specimen remaining the in **cobas**® PCR Media tube.

Endocervical and vaginal specimens

- The presence of mucus in endocervical specimens may cause processing delays due to clotting. Mucus free specimens are required for optimal test performance. Use the large woven polyester swab in the **cobas**® PCR Dual Swab Sample Kit or an equivalent device to remove cervical secretions and discharge before obtaining the endocervical specimen.
- Use only the flocked swab in the **cobas**® PCR Media Dual Swab Sample Kit to collect endocervical specimens. Use only the woven polyester swab in either the **cobas**® PCR Media Uni Swab Sample Kit or the **cobas**® PCR Media Dual Swab Sample Kit to collect vaginal swab specimens. **cobas**® CT/NG has not been validated for use with other swab collection devices or media types. Using **cobas**® CT/NG with other swab collection devices or media types may lead to false negative, false positive, and/or invalid results.
- To avoid cross contamination of processed specimens, additional caps for **cobas**® PCR Media tubes in an alternate color (neutral; see **Additional materials required**) should be used to recap specimens after processing.
- All swab specimens containing a single swab in the **cobas**® PCR Media tube can be directly processed on the **cobas**® 6800/8800 Systems. If desired, the swab may be removed before the specimen tube is loaded onto the instrument, however utmost care must be exercised to avoid cross contamination.
- A properly collected swab specimen should have a single swab with the shaft broken at the score line. Swab shafts which are broken above the score line will appear longer than normal and may also be bent over to fit into the **cobas**® PCR Media tube. This can create an obstruction to the pipetting system which may cause the loss of sample, test results and/or mechanical damage to the instrument. In the event that a swab specimen has an improperly broken shaft, remove the swab prior to sample processing on the **cobas**® 6800/8800 Systems. Use caution when disposing of specimen swabs; avoid splashing or touching swabs to other surfaces during disposal to prevent contamination.
- Incoming primary swab specimen tubes with no swabs or with multiple swabs have not been collected according to the instructions in their respective collective kit IFU and should not be tested.
- Occasionally, incoming swab specimens contain excessive mucus which may induce a pipetting error (e.g., clot or other obstruction) on the **cobas**® 6800/8800 Systems. Prior to retesting of specimens that exhibited clots during initial processing, remove and discard the swab, then re-cap and vortex these specimens for 30 seconds to disperse the excess mucus.
- Swab specimens can be assayed twice on the **cobas**® 6800/8800 Systems while the swab is in the collection tube. If additional testing is required, or if the first test fails due to specimen pipetting error (e.g., clot or other obstruction), the swab must be removed prior to testing and the remaining fluid must have a minimum volume of 1.0 mL.

Cervical specimens in PreservCyt® Solution

- **cobas**® CT/NG is validated for use with cervical specimens collected in PreservCyt® Solution prior to cytology processing. **cobas**® CT/NG has not been validated for use with cervical specimens obtained in other media types. Using **cobas**® CT/NG with other media types may lead to false negative, false positive, and/or invalid results.
- Cervical specimens in PreservCyt® Solution should be aliquoted into secondary tubes as follows:
    1. Prepare a barcoded 13 mL round-bottom **cobas**® PCR Secondary tube (*see **Additional materials required***) for each PreservCyt® specimen to be tested.
    2. With clean gloved hands, **vortex** each PreservCyt® primary specimen vial for **10 seconds** immediately **prior** to transfer.
    3. Uncap a primary vial and transfer at least **1.0 mL** but no more than **4.0 mL** into the prepared barcoded secondary tube from step 1.
        - *Always use caution when transferring specimens from primary containers to secondary tubes.*
        - *Always use a new pipette tip for each specimen.*
        - *Always use pipettors with aerosol-barrier or positive-displacement tips to handle specimens.*
        - *To avoid cross contamination, additional caps for these tubes in an alternate color (neutral; see **Additional materials required**) should be used to recap these specimens after processing.*
        - *Transfer tube to a rack if testing is to be performed shortly after or cap the secondary tube if testing will be performed at a future time.*
    4. Re-cap the primary vial with a replacement cap before moving to the next specimen. Store the primary vial upright.
    5. Only racks of uncapped tubes may be loaded into the Sample Supply Module of the **cobas**® 6800/8800 Systems for CT/NG testing.
- Cervical specimens in PreservCyt® Solution can be assayed twice on the **cobas**® 6800/8800 Systems as long as the minimum volume requirements are met.

Instructions for use

Procedural notes

- Do not use **cobas**® CT/NG, **cobas**® CT/NG Positive Control Kit, **cobas**® Buffer Negative Control Kit, or **cobas omni** reagents after their expiry dates.
- Do not reuse consumables. They are for one-time use only.
- Ensure that specimen barcode labels on sample tubes are visible through the openings on the side of MPA sample racks. Refer to the **cobas**® 6800/8800 Systems User Guide for proper barcode specifications and additional information on loading sample tubes.
- Refer to the **cobas**® 6800/8800 Systems User Guide for proper maintenance of instruments.

Running cobas® CT/NG

cobas® CT/NG can be run with a minimum required sample volume of 1.0 mL for swab and PreservCyt® specimens, and 1.2 mL for urine specimens. The operation of the instrument is described in detail in the **cobas**® 6800/8800 Systems User Guide. Figure 1 below summarizes the procedure.

- Swab and urine specimens must be uncapped and loaded directly onto racks for processing on the **cobas**® 6800/8800 Systems.
- It is necessary to aliquot specimens collected in PreservCyt® Solution. Refer to the preparation instructions for cervical specimens found in section: "Cervical specimens in PreservCyt® Solution".
- A single run can have any combination of specimens (Swab, Urine, and PreservCyt®) and each specimen can be tested with either the CT/NG, CT, or NG ASAPs.
- Specimens collected in **cobas**® PCR Media or PreservCyt® Solution should be processed using the sample type selection in the user interface (UI) of the **cobas**® CT/NG as described in Table 11.

Table 11 Sample type selection in the user interface of the **cobas**® CT/NG

Specimen		**Collection kit type**	**Process as Sample Type**
Female	Vaginal swab	**cobas**® PCR Media Uni or Dual Swab Sample Kit	**Swab**
	Endocervical swab	**cobas**® PCR Media Dual Swab Sample Kit	**Swab**
	Urine	**cobas**® PCR Urine Sample Kit	**Urine**
	Cervical specimen	PreservCyt® Solution (ThinPrep)	**PreservCyt®**
Male	Urine	**cobas**® PCR Urine Sample Kit	**Urine**

Figure 1 **cobas**® CT/NG procedure

1

Log onto the system
Press Start to prepare the system
Order Tests
- Choose “Swab” for ordering swab specimens collected in **cobas**® PCR Media
- Choose “Urine” for ordering urine specimens collected in **cobas**® PCR Media
- Choose “PreservCyt” for ordering PreservCyt® Solution specimens

Refill reagents and consumables as prompted by the system
- Load test specific reagent cassette
- Load control cassettes
- Load pipette tips
- Load processing plates
- Load MGP Reagent
- Load amplification plates
- Refill Specimen Diluent
- Refill Lysis Reagent
- Refill Wash Reagent

3

Loading specimens onto the system
- For each primary urine or swab in **cobas**® PCR Media
  - Uncap tube
  - Transfer tube directly to rack
- For each primary PreservCyt® specimen vial:
  - Vortex for 10 seconds
  - Aliquot a minimum of 1 mL of PreservCyt® specimen into a 13 mL round-bottom secondary tube
  - Transfer tube to rack
- Load sample rack and clot tip racks into the sample supply module
- Confirm samples have been accepted into the transfer module

4

Start run

5

Review and export results

6

Remove sample tubes. If needed, cap any sample tubes meeting the minimum volume requirements for future use.

Clean up instrument
- Unload empty control cassettes
- Empty amplification plate drawer
- Empty liquid waste
- Empty solid waste

Results

cobas® CT/NG automatically detects and discriminates CT and/or NG DNA simultaneously for each individually processed sample and control, displaying individual target results for samples as well as validity and overall results for controls.

Quality control and validity of results

- One **cobas**® Buffer Negative Control [(-) Ctrl] and one CT/NG Positive Control [CT/NG (+) C] are processed with each batch of a requested result type.
- In the **cobas**® 6800/8800 software and/or report, check for flags and their associated results to ensure batch validity.
- All flags are described in the cobas® 6800/8800 Systems User Guide.
- The batch is valid if no flags appear for any controls. If the batch is invalid, repeat testing of the entire batch.

Validation of results is performed automatically by the **cobas**® 6800/8800 software based on negative and positive control performance.

Interpretation of results

Display examples for **cobas**® CT/NG are shown in Figure 2, Figure 3, and Figure 4, respectively.

Figure 2 Example of **cobas**® CT/NG results display for the CT/NG result request

Test	Sample ID	Valid	Flags	Sample type	Overall result	Target 1	Target 2
CT/NG	C161420284084196207422	Yes		CT/NG (+) C	**Valid**	Valid	Valid
CT/NG	C161420284090419545972	Yes		(-) Ctrl	**Valid**	Valid	Valid
CT/NG 400 ul	CTNG_PC1	NA		PreservCyt®	**NA**	CT Positive	NG Positive
CT/NG 400 ul	CTNG_PC2	NA		PreservCyt®	**NA**	CT Negative	NG Positive
CT/NG 400 ul	CTNG_Swab1	NA		Swab	**NA**	CT Negative	NG Negative
CT/NG 400 ul	CTNG_Swab2	NA		Swab	**NA**	CT Positive	NG Positive
CT/NG 400 ul	CTNG_Swab3	NA	C02H2	Swab	**NA**	CT Positive	Invalid
CT/NG 850 ul	CTNG_Urine1	NA		Urine	**NA**	CT Positive	NG Negative
CT/NG 850 ul	CTNG_Urine2	NA		Urine	**NA**	CT Negative	NG Negative
CT/NG 850 ul	CTNG_Urine3	NA	Y40T	Urine	**NA**	Invalid	Invalid

Figure 3 Example of **cobas**® CT/NG results display for the CT result request

Test	Sample ID	Valid	Flags	Sample type	Overall result	Target 1	Target 2
CT	C161420284084196207423	Yes		CT/NG (+) C	**Valid**	Valid	
CT	C161420284090419545973	Yes		(-) Ctrl	**Valid**	Valid	
CT 400 ul	CT_PC1	NA		PreservCyt®	**NA**	CT Positive	
CT 400 ul	CT_PC2	NA		PreservCyt®	**NA**	CT Positive	
CT 400 ul	CT_Swab1	NA		Swab	**NA**	CT Negative	
CT 400 ul	CT_Swab2	NA		Swab	**NA**	CT Positive	
CT 400 ul	CT_Swab3	NA	P02T	Swab	**NA**	Invalid	
CT 850 ul	CT_Urine1	NA		Urine	**NA**	CT Negative	
CT 850 ul	CT_Urine2	NA		Urine	**NA**	CT Positive	

Note: The Target 2 column is reserved for NG results.

Figure 4 Example of **cobas**® CT/NG results display for the NG result request

Test	Sample ID	Valid	Flags	Sample type	Overall result	Target 1	Target 2
NG	C161420284084196207424	Yes		CT/NG (+) C	**Valid**		Valid
NG	C161420284090419545974	Yes		(-) Ctrl	**Valid**		Valid
NG 400 ul	NG_PC1	NA		PreservCyt®	**NA**		NG Negative
NG 400 ul	NG_PC2	NA		PreservCyt®	**NA**		NG Positive
NG 400 ul	NG_PC3	NA	Y40T	PreservCyt®	**NA**		Invalid
NG 400 ul	NG_Swab1	NA		Swab	**NA**		NG Positive
NG 400 ul	NG_Swab2	NA		Swab	**NA**		NG Negative
NG 850 ul	NG_Urine1	NA		Urine	**NA**		NG Negative
NG 850 ul	NG_Urine2	NA		Urine	**NA**		NG Positive

Note: The Target 1 column is reserved for CT results.

For a valid batch, check each individual sample for flags in the **cobas**® 6800/8800 software and/or report. The result interpretation should be as follows:

- A valid batch may include both valid and invalid sample results.
- The "Valid" and "Overall Result" columns are not applicable (NA) to sample results for **cobas**® CT/NG and are marked with "NA". Values reported in these columns **do not** impact the validity of results reported within individual Target Result columns.
- Reported target results for individual samples are valid unless indicated as "Invalid" within the individual target result column.
- Invalid results for one or more target combinations are possible with the CT/NG result request and are reported out specifically for each channel. Refer to retesting instructions as described in Table 12 through Table 14, and within the "Specimen collection, transport, and storage" section for additional specimen type specific information.
- Results of this test should only be interpreted in conjunction with information available from clinical evaluation of the patient and patient history.

Results and their corresponding interpretation for detecting CT and NG (Table 12), CT only (Table 13) and NG only (Table 14) are shown below.

Table 12 cobas® CT/NG results and interpretation for the CT/NG result request

Target 1	Target 2	Interpretation
CT Positive	NG Positive	All requested results were valid. Target signal detected for CT and NG DNA.
CT Positive	NG Negative	All requested results were valid. Target signal detected for CT DNA. No target signal detected for NG DNA.
CT Negative	NG Positive	All requested results were valid. No target signal detected for CT DNA. Target signal detected for NG DNA.
CT Negative	NG Negative	All requested results were valid. No target signal detected for CT or NG DNA.
CT Positive	Invalid	Not all requested results were valid. CT result is valid. Target signal detected for CT DNA. NG result is invalid. Original specimen should be re-tested to obtain valid NG results. If the result is still invalid, a new specimen should be obtained.
Invalid	NG Positive	Not all requested results were valid. CT result is invalid. Original specimen should be re-tested to obtain valid CT results. If the result is still invalid, a new specimen should be obtained. NG result is valid. Target signal detected for NG DNA.
CT Negative	Invalid	Not all requested results were valid. CT result is valid. No target signal detected for CT DNA. NG result is invalid. Original specimen should be re-tested to obtain valid NG results. If the result is still invalid, a new specimen should be obtained.
Invalid	NG Negative	Not all requested results were valid. CT result is invalid. Original specimen should be re-tested to obtain valid CT results. If the result is still invalid, a new specimen should be obtained. NG result is valid. No target signal detected for NG DNA.
Invalid	Invalid	Both CT and NG results are invalid. Original specimen should be re-tested to obtain valid CT and NG results. If the results are still invalid, a new specimen should be obtained.

Table 13 cobas® CT/NG results and interpretation for the CT result request

Target 1	Target 2	Interpretation
CT Positive	<Blank>	The requested result was valid. Target signal detected for CT DNA.
CT Negative	<Blank>	The requested result was valid. No target signal detected for CT DNA
Invalid	<Blank>	CT result is invalid. Original specimen should be re-tested to obtain valid CT results. If the result is still invalid, a new specimen should be obtained.

Table 14 cobas® CT/NG results and interpretation for the NG result request

Target 1	Target 2	Interpretation
<Blank>	NG Positive	The requested result was valid. Target signal detected for NG DNA.
<Blank>	NG Negative	The requested result was valid. No target signal detected for NG DNA
<Blank>	Invalid	NG result is invalid. Original specimen should be re-tested to obtain valid NG results. If the result is still invalid, a new specimen should be obtained.

Procedural limitations

- **cobas**® CT/NG has been evaluated only for use in combination with the **cobas**® CT/NG Positive Control Kit, **cobas**® Buffer Negative Control Kit, **cobas omni** MGP Reagent, **cobas omni** Lysis Reagent, **cobas omni** Specimen Diluent, and **cobas omni** Wash Reagent for use on the **cobas**® 6800/8800 Systems.
- Reliable results depend on proper sample collection, storage and handling procedures.
- **cobas**® CT/NG has only been validated for use with male and female urine, clinician-instructed self-collected vaginal swab specimens, clinician-collected vaginal swab specimens, and endocervical swab specimens, all collected in **cobas**® PCR Media (Roche Molecular Systems, Inc.) and cervical specimens collected in PreservCyt® Solution. Assay performance has not been validated for use with other collection media and/or specimen types.
- Detection of *C. trachomatis* and *N. gonorrhoeae* is dependent on the number of organisms present in the specimen and may be affected by specimen collection methods, patient factors (i.e., age, history of STD, presence of symptoms), stage of infection and/or infecting *C. trachomatis* and *N. gonorrhoeae* strains.
- Though rare, mutations within the highly conserved regions of the cryptic plasmid or genomic DNA of *C. trachomatis* or the genomic DNA of *N. gonorrhoeae* covered by **cobas**® CT/NG primers and/or probes may result in failure to detect the presence of the bacterium.
- Due to inherent differences between technologies, it is recommended that, prior to switching from one technology to the next, users perform method correlation studies in their laboratory to qualify technology differences. One hundred percent agreement between the results should not be expected due to aforementioned differences between technologies. Users should follow their own specific policies/procedures.
- **cobas**® CT/NG is not intended to replace other exams or tests for diagnosis of urogenital infection. Patients may have cervicitis, urethritis, urinary tract infections, or vaginal infections due to other causes or concurrent

infections with other agents.

- **cobas**® CT/NG is not recommended for evaluation of suspected sexual abuse and for other medico-legal indications.
- **cobas**® CT/NG should not be used to determine therapeutic success as nucleic acids may be present after antimicrobial therapy.

- **cobas**® CT/NG for urine testing is recommended to be performed on first catch urine specimens (defined as the first 10 to 50 mL of the urine stream). The effects of other variables such as first-catch vs. mid-stream, post-douching, etc. have not been evaluated.
- The effects of other potential variables such as vaginal discharge, use of tampons, douching, etc. and specimen collection variables have not been evaluated.
- **cobas**® CT/NG has not been evaluated with patients who are currently being treated with antimicrobial agents active against CT or NG as well as patients with a history of hysterectomy.
- False negative or invalid results may occur due to polymerase inhibition. The CT/NG Internal Control is included in **cobas**® CT/NG to help identify the specimens containing substances that may interfere with nucleic acid isolation and PCR amplification.
- The addition of AmpErase enzyme into the **cobas**® CT/NG Master Mix reagent enables selective amplification of target DNA; however, good laboratory practices and careful adherence to the procedures specified in this Package Insert are necessary to avoid contamination of reagents.
- **cobas**® CT/NG has not been evaluated in patients younger than 14 years of age.
- Urogenital specimens from patients who have used the over-the-counter products Replens™ Long-Lasting Vaginal Moisturizer, RepHresh™ Odor Eliminating Vaginal Gel and RepHresh™ Clean Balance or used Metronidazole Vaginal Gel may generate invalid or false negative results. See Interference results (Table 22) for further details.
- The presence of mucus (> 0.5% w/v) in endocervical specimens may cause false negative test results.
- The presence of whole blood (> 5% v/v) in urine and cervical specimens collected in PreservCyt® Solution may cause false negative and/or invalid test results. Do not test specimens that appear bloody or have a dark brown color.
- When *C.trachomatis* is present at very high concentration, ($\geq 10^3$ IFU/mL, corresponding to less than 5% of positive clinical samples), the detection of *N.gonorrhoeae* present at concentrations near the limit of detection (LoD) of **cobas**® CT/NG may be impacted.

Non-clinical performance evaluation

Analytical sensitivity (Limit of Detection)

Analytical sensitivity (Limit of Detection or LoD) was determined by analyzing a dilution series of quantified cultures of *Chlamydia trachomatis* (serovars D and I) and *Neisseria gonorrhoeae* isolates 2948 (ATCC 19424) and 891. CT and NG cultures were diluted into a matrix of pooled negative specimens of each sample type and 70-78 replicates were tested for each level in each specimen type. All levels were analyzed across 3 unique lots of reagents. LoD for each specimen type is shown in Table 15 as the target concentration which can be detected in ≥ 95% of the replicates for all lots.

Table 15 Analytical sensitivity (Limit of Detection)

Specimen Types	*C. trachomatis*				*N. gonorrhoeae*			
	Serovar D		Serovar I		Strain 2948		Strain 891	
	LoD (IFU/mL)	Mean Ct Value	LoD (IFU/mL)	Mean Ct Value	LoD (CFU/mL)	Mean Ct Value	LoD (CFU/mL)	Mean Ct Value
Endocervical Swab in **cobas**® PCR Media	0.3	36.6	1.4	37.1	0.4	36.3	0.08	37.5
Vaginal Swab in **cobas**® PCR	0.3	37.3	1.4	37.0	0.4	36.3	0.08	37.0
cobas® PCR Media	0.2	37.8	1.3	37.1	0.2	36.3	0.04	38.3
Cervical Samples collected into PreservCyt® Solution	0.6	37.4	2.9	37.4	0.2	36.7	0.08	37.5

IFU = Inclusion Forming Unit; quantification of the same *C. trachomatis* culture using DFA method equates 1 IFU to 6.6 signal generating units (SGU) for Serovar D, and 13.9 SGU for serovar I, where SGU includes Elementary Bodies as well as Reticulate Bodies of *C. trachomatis*

CFU = Colony Forming Units

Inclusivity

Inclusivity and verification of the LoD were performed for 13 additional CT serovars, the Swedish new variant strain (nvCT) and an additional 43 independently isolated strains of NG using one lot of reagents. Testing was performed using CT and NG cultures diluted into pools of negative specimens. Results are shown in Table 16 and Table 17 for CT serovars and NG strains, respectively. Twenty replicates per dilution level were tested for each strain in each specimen type.

Table 16 Inclusivity testing for CT serovars

Serovar Type or Variant	Swab* Specimens		Urine Specimens		PreservCyt® Specimens	
	IFU/mL	% Pos	IFU/mL	% Pos	IFU/mL	% Pos
A	1.4	100	0.7	100	1.4	100
B	5.9	100	2.9	100	5.9	100
Ba	18.3	100	9.1	100	18.3	100
C	0.6	100	0.3	100	0.6	100
E	6.4	100	3.2	100	6.4	100
F	3.2	100	1.6	100	3.2	100
G	2.9	100	1.5	100	2.9	100
H	9.7	100	4.8	100	9.7	100
J	1.4	100	0.7	100	1.4	100
K	2.0	100	1.0	100	2.0	100
LGV Type 1	5.9	100	3.0	100	5.9	100
LGV Type 2	12.8	100	6.4	100	12.8	100
LGV Type 3	0.7	100	0.4	100	0.7	100
nvCT	0.7	100	0.3	100	0.7	100

* Vaginal swab samples were used as a representative swab sample type for vaginal and endocervical swab specimens.

Table 17 Inclusivity testing for NG strains

Numbers of NG Strains	Swab* Specimens	
	CFU/mL	% Pos
39	0.4	≥ 95
4	1.0	≥ 95
Total = 43		
Numbers of NG Strains	**Urine Specimens**	
	CFU/mL	**% Pos**
41	0.2	≥ 95
2	0.5	100
Total = 43		
Numbers of NG Strains	**PreservCyt® Specimens**	
	CFU/mL	**% Pos**
42	0.4	≥ 95
1	1.0	100
Total = 43		

* Vaginal swab samples were used as a representative swab sample type for vaginal and endocervical swab specimens.

Precision (within laboratory)

In-house precision was examined using a panel composed of CT and NG cultures diluted into a pool of negative endocervical swab specimen matrix collected in **cobas**® PCR Media, a pool of negative urine matrix plus **cobas**® PCR Media and a pool of negative cervical specimen matrix collected in PreservCyt® Solution. Endocervical swabs were intended to represent swab samples collected in **cobas**® PCR Media (endocervical and vaginal). Four levels were tested using CT serovar D and NG strain 2948 (ATCC 19424) as the target organisms.

The precision panel was designed to include members with high negative, low and moderate concentrations of CT and NG for each panel matrix, corresponding to ~0.3x, ~1x and ~3x LoD. Testing was performed with three lots of **cobas**® CT/NG reagents and two instruments for a total of 24 runs. A description of the precision panels and the study performance hit rate is shown in Table 18. All negative panel members tested negative throughout the study. Analysis of standard deviation and percent coefficient of variation of the Ct values from valid tests performed on positive panel members (see Table 19 and Table 20) yielded overall CV (%) ranges from 1.62% to 4.05% for CT and from 1.17% to 3.55% for NG. Testing occurred over 12 days, using 2 instruments, with 2 runs per day. Each run consisted of 3 replicates of each sample.

Table 18 Summary of within-laboratory precision

Level	N Tested	N positive CT	N positive NG	Hit Rate		95% CI CT		95% CI NG	
				CT	NG	LL	UL	LL	UL
Endocervical Swab in **cobas**® PCR Media									
Negative	72	0	0	0%	0%	0.0	5.0	0.0	5.0
High Negative	72	51	32	71%	44%	59	81	33	57
Low	72	69	68	96%	94%	88	99	86	98
Moderate	72	72	72	100%	100%	95	100	95	100
Cervical samples collected into PreservCyt® Solution									
Negative	72	0	0	0%	0%	0.0	5.0	0.0	5.0
High Negative	72	38	47	53%	65%	41	65	53	76
Low	72	72	69	100%	96%	95	100	88	99
Moderate	72	72	72	100%	100%	95	100	95	100
cobas® PCR Media with Urine									
Negative	72	0	0	0%	0%	0.0	5.0	0.0	5.0
High Negative	72	56	56	78%	78%	66	87	66	87
Low	72	71	72	99%	100%	92	100	95	100

Moderate	72	72	72	100%	100%	95	100	95	100

Table 19 Overall mean, standard deviations and coefficients of variation (%) for cycle threshold, CT positive panel members

Level (Hit Rate)	Mean Ct	Between instrument		Between lot		Within run		Between run		Between day		Total	
		SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Endocervical Swab in **cobas**® PCR Media													
High Negative (71%)	39.7	0.00	0.00	0.00	0.00	1.27	3.21	0.00	0.00	0.34	0.85	1.32	3.32
Low (96%)	38.5	0.00	0.00	0.04	0.10	1.14	2.96	0.00	0.00	0.48	1.25	1.24	3.22
Moderate (100%)	36.9	0.00	0.00	0.25	0.69	0.54	1.45	0.07	0.18	0.00	0.00	0.60	1.62
Cervical Samples collected into PreservCyt® Solution													
High Negative (53%)	38.3	0.60	1.57	0.52	1.37	1.12	2.92	0.00	0.00	0.00	0.00	1.37	3.58
Low (100%)	36.9	0.21	0.56	0.28	0.76	0.68	1.85	0.00	0.00	0.00	0.00	0.77	2.08
Moderate (100%)	35.6	0.00	0.00	0.20	0.56	0.52	1.46	0.09	0.24	0.02	0.05	0.56	1.59
cobas® PCR Media with Urine													
High Negative (78%)	38.9	0.00	0.00	0.12	0.30	1.25	3.22	0.39	1.01	0.00	0.00	1.32	3.39
Low (99%)	38.3	0.11	0.28	0.00	0.00	1.52	3.97	0.00	0.00	0.29	0.77	1.55	4.05
Moderate (100%)	37.1	0.00	0.00	0.00	0.00	1.05	2.84	0.00	0.00	0.28	0.77	1.09	2.94

Table 20 Overall mean, standard deviations and coefficients of variation (%) for cycle threshold, NG positive panel members

Level (Hit Rate)	Mean Ct	Between instrument		Between lot		Within run		Between run		Between day		Total	
		SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%
Endocervical Swab in **cobas**® PCR Media													
High Negative (44%)	39.1	0.00	0.00	0.31	0.79	0.84	2.14	0.72	1.85	0.57	1.46	1.28	3.28
Low (94%)	38.1	0.00	0.00	0.00	0.00	1.27	3.34	0.00	0.00	0.00	0.00	1.27	3.34
Moderate (100%)	36.5	0.00	0.00	0.24	0.67	0.69	1.89	0.00	0.00	0.15	0.40	0.74	2.04
Cervical Samples collected into PreservCyt® Solution													
High Negative (65%)	39.0	0.34	0.87	0.00	0.00	1.11	2.85	0.08	0.20	0.45	1.16	1.25	3.21
Low (96%)	38.0	0.00	0.00	0.00	0.00	1.25	3.28	0.00	0.00	0.00	0.00	1.25	3.28
Moderate (100%)	35.8	0.00	0.00	0.28	0.78	0.76	2.13	0.00	0.00	0.00	0.00	0.81	2.27
cobas® PCR Media with Urine													
High Negative (78%)	39.1	0.00	0.00	0.26	0.66	1.35	3.46	0.00	0.00	0.18	0.45	1.39	3.55
Low (100%)	36.7	0.14	0.38	0.16	0.42	0.71	1.92	0.00	0.00	0.00	0.00	0.74	2.00
Moderate (100%)	34.9	0.00	0.00	0.16	0.47	0.37	1.06	0.06	0.18	0.00	0.00	0.41	1.17

Analytical specificity/cross-reactivity

A panel of 149 bacteria, fungi and viruses, including those commonly found in the male and female urogenital tract, 20 representatives of non-*gonorrhoeae Neisseria* strains and other phylogenetically unrelated organisms, were tested with **cobas**® CT/NG to assess analytical specificity. The organisms listed in Table 21 were spiked at concentrations of approximately 1 x 10^6 units*/mL for bacteria and approximately 1 x 10^5 units*/mL for viruses into pools of negative vaginal swab specimens in **cobas**® PCR Media, urine stabilized in **cobas**® PCR Media and cervical specimens in PreservCyt® Solution. Testing was performed with each potential interfering organism alone as well as with each organism mixed with CT and NG cultures at ~3x LoD. Results indicated that none of these organisms interfered with the detection of CT and NG or produced false positive results in the CT/NG negative matrices (N=3 across the tested specimen types).

*All bacteria were quantified as Colony Forming Units (CFU) except *Chlamydophila pneumonia* and *Chlamydophila psittaci* which were quantified as Elementary Bodies (EB). All viruses were quantified as units/mL as determined by

$TCID_{50}$ Endpoint Dilution Assay. *Trichomonas vaginalis* and HPV16 were quantified as cells/mL.

Table 21 Microorganisms tested for analytical specificity/cross reactivity

Achromobacter xerosis	*Gemella haemolysans*	*Neisseria subflava*
Acinetobacter calcoaceticus	*Haemophilus ducreyi*	*Neisseria weaverii*
Acinetobacter lwoffi	*Haemophilus influenzae*	*Pantoea agglomerans*
Actinomyces israelii	*Helicobacter pylori*	*Paracoccus denitrificans*
Aerococcus viridans	Herpes simplex virus I	*Peptostreptococcus anaerobius*
Aeromonas hydrophila	Herpes simplex virus II**	*Peptostreptococcus asaccharolyticus*
Alcaligenes faecalis	HPV16*	*Peptostreptococcus magnus*
Atopobium vaginae	*Kingella dentrificans*	*Plesiomonas shigelloides*
Bacillus subtilis	*Kingella kingae*	*Propionibacterium acnes*
Bacteriodes fragilis	*Klebsiella oxytoca*	*Proteus mirabilis*
Bacteroides caccae	*Klebsiella pneumoniae*	*Proteus penneri*
Bacteroides ureolyticus	*Lactobacillus acidophillus*	*Proteus vulgaris*
Bergeriella denitrificans	*Lactobacillus brevis*	*Providencia rettgeri*
Bifidobacterium adolescentis	*Lactobacillus crispatus*	*Providencia stuartii*
Bifidobacterium breve	*Lactobacillus jensenii*	*Pseudomonas aeruginosa*
Bifidobacterium longum	*Lactobacillus lactis*	*Pseudomonas fluorescens*
Blautia product	*Lactobacillus leichmannii*	*Pseudomonas putida*
Branhamella catarrhalis	*Lactobacillus oris*	*Rahnella aquatilis*
Brevibacterium linens	*Lactobacillus parabuchnerri*	*Rhizobium radiobacter*
Campylobacter coli	*Lactobacillus reuteri*	*Rhodospirillum rubrum*
Campylobacter jejuni	*Lactobacillus vaginalis*	*Saccharomyces cerevisiae*
Candida albicans	*Lactococcus lactis cremoris*	*Salmonella choleraesuis*
Candida glabrata	*Legionella pneumophila*	*Salmonella minnesota*
Candida parapsilosis	*Leuconostoc paramesenteroides*	*Salmonella typhimurium*
Candida tropicalis	*Listeria monocytogenes*	*Serratia denitrificans*
Chlamydophila pneumoniae	*Micrococcus luteus*	*Serratia marcescens*
Chlamydophila psittaci	*Moraxella lacunata*	*Shigella dysenteriae*
Chromobacter violaceum	*Moraxella osloensis*	*Staphylococcus aureus*
Citrobacter freundii	*Morganella morganii*	*Staphylococcus epidermidis*
Clostridium difficile	*Mycobacterium smegmatis*	*Staphylococcus saprophyticus*
Clostridium perfringens	*Mycoplasma genitalium***	*Streptococcus agalactiae*
Corynebacterium genitalium	*Mycoplasma hominis*	*Streptococcus anginosus*
Corynebacterium xerosis	*Neisseria cinerea*	*Streptococcus bovis*
Cryptococcus neoformans	*Neisseria elongata subsp. elongata*	*Streptococcus dysgalactiae*
*Cytomegalovirus***	*Neisseria elongata subsp. nitroreducens*	*Streptococcus equinis*
Deinococcus radiodurans	*Neisseria flava*	*Streptococcus mitis*
Derxia gummosa	*Neisseria flavescens*	*Streptococcus mutans*
Eikenella corrodens	*Neisseria kochi*	*Streptococcus pneumoniae*
Enterobacter aerogenes	*Neisseria lactamica*	*Streptococcus pyogenes*
Enterobacter cloacae	*Neisseria macacae*	*Streptococcus salivarius*
Enterococcus avium	*Neisseria meningitidis Serogroup A*	*Streptococcus sanguis*
Enterococcus casseliflavus	*Neisseria meningitidis Serogroup B*	*Streptomyces griseinus*

Enterococcus faecalis	*Neisseria meningitidis Serogroup C*	*Trichomonas vaginalis*
Enterococcus faecium	*Neisseria meningitidis Serogroup D*	*Trueperella pyogenes*
Erysipelothrix rhusiopathiae	*Neisseria meningitidis Serogroup W135*	*Ureaplasma urealyticum*
Escherichia coli	*Neisseria meningitidis Serogroup Y*	*Veillonela parvula*
Escherichia fergusonii	*Neisseria mucosa*	*Vibrio cholerae*
Flavobacterium meningosepticum	*Neisseria perflava*	*Vibrio parahaemolyticus*
Fusobacterium nucleatum	*Neisseria polysaccharea*	*Yersinia enterocolitica*
Gardnerella vaginalis	*Neisseria sicca*	-

* HPV16 was tested as CaSki cells.

** Organism was tested at a concentration of 1 x 10^4 Units/mL.

***Organism was tested at a concentration of 1 x 10^5 CFU/mL.

Interference

The effects of over-the-counter or prescription products that may be present in urogenital specimens (Table 22), were evaluated. Testing was done using pooled clinical specimens (vaginal swab, urine and PreservCyt® specimens) with spiking of potential interferents at levels expected from normal patient usage. Interferents were tested in CT/NG negative specimen pools as well as in specimen pools with CT/NG at ~3x LoD in the specimen type tested. CT serovars D and I and NG strains 2948 (ATCC 19424) and 891 were used in this study. Five replicates each of CT/NG negative and CT/NG positive samples were tested with each product in each specimen type, except for RepHresh™ Odor Eliminating Vaginal Gel and RepHresh™ Clean Balance Gel, which were tested with 2 replicates each to verify interference that had been observed with Replens™ Long-Lasting Vaginal Moisturizer, a product with a similar formulation.

Of the over-the-counter (OTC) and prescription products tested, Metronidazole Vaginal Gel, Replens™ Long-Lasting Vaginal Moisturizer, RepHresh™ Odor Eliminating Vaginal Gel and RepHresh™ Clean Balance produced false negative or invalid results in at least one replicate of the samples tested.

Table 22 List of substances with concentrations that do not interfere with test performance in urogenital specimens

Product Name	Vaginal Swabs	Urine	PreservCyt® Solution
	mg/mL	mg/mL	mg/mL
Clindamycin Phosphate Vaginal Cream	7.1	3.4	1.6
Equate tioconazole 1	3.7	1.7	0.8
Equate Vagicaine Anti-Itch Cream	4.1	2.0	0.9
Estrace	3.8	2.0	1.0
K-Y™ Ultra Gel	5.7	2.7	1.2
Metronidazole Vaginal Gel	0.1*	0.1*	0.2*
Monistat 3 Vaginal Antifungal Combination Pack	3.7	1.7	0.7
Monistat® Complete Care Itch Relief Cream	3.7	1.8	0.9
7 Day Vaginal Cream	3.9	1.8	0.8
Norforms Suppositories	3.4	1.7	0.7
Premarin	6.1	3.1	1.4
Replens™ Long-Lasting Vaginal Moisturizer	0.05*	0.05*	0.2*
Summer's Eve Feminine Deodorant Spray	6.4	3.1	2.0
VCF - Vaginal Contraceptive Foam	2.1	1.0	0.4
Yeast Gard Advanced	3.7	1.7	1.0
Azo Standard (urine only)	N/A	0.1	N/A
RepHresh™ Odor Eliminating Vaginal Gel	‡	‡	‡
RepHresh™ Clean Balance Gel	‡	‡	‡

* Concentrations above this level may cause interference in clinical samples

‡ RepHresh™ products were tested using simulated swab specimen. Concentrations of product that did not interfere with test performance were not determined

Endogenous substances that may be present in urogenital specimens were tested for interference. Testing was done using pooled clinical specimens (endocervical swab, urine and PreservCyt® specimens) with spiking of potential endogenous interferents. Interferents were tested in CT/NG negative specimen pools as well as in the presence of CT/NG at ~3x LoD in the specimen type tested. CT serovars D and I and NG strains 2948 (ATCC 19424) and 891 were used in this study. Five replicates each of CT/NG negative and CT/NG positive samples were tested with each substance in each specimen type.

Interference was noted with whole blood at 10% for urine and PreservCyt® specimens and with cervical mucus at 1% in endocervical specimens when at least one replicate of the samples tested produced false negative or invalid results. Levels of endogenous substances tolerated by the assay for all specimen types are shown in Table 23.

Table 23 Summary of endogenous substance concentrations that do not show interference

Interferent	Endocervical Swab	PreservCyt®	Urine
Albumin (% w/v)	N/A	N/A	5%
Bilirubin (% w/v)	N/A	N/A	0.5%
Mucus (% w/v)	0.5%	1.0%	0.5%
Glucose (% w/v)	N/A	N/A	1.0%
Peripheral Blood Mononuclear Cells (PBMCs as cells/mL)	1.0E+06	1.0E+06	1.0E+06
pH (acidic and alkaline)	N/A	N/A	pH 4 and pH 9
Semen (% w/v)	1.5%	1.5%	N/A
Whole Blood (% v/v)	10%	5%	5%

Competitive inhibition

To assess competitive inhibition between CT and NG, samples of vaginal swab, urine and PreservCyt® specimens were tested with low and moderate concentrations of one target mixed with very high concentrations of the opposite target. Low and moderate concentrations were defined as ~1x LoD and ~3x LoD, respectively, and high concentrations ($\geq 10^3$ IFU/mL for CT and $\geq 10^4$ CFU/mL for NG) were defined as generating a signal greater than observed in 95% of target positive clinical specimens.

Testing results indicated that when NG was present at a high concentration, CT was detected in all specimen types, at both low (~1x LoD) and moderate (~3x LoD) levels. Results also indicated that when CT was present at a high concentration, NG was detected in all specimen types at moderate (~3x LoD) levels, however, NG was not consistently detected at low levels (Expanded testing indicated detection in 35% (7/20) of the samples at 0.4 CFU/ml and 60% (12/20) of the samples at 0.65 CFU/mL).

Cross-contamination/Carryover

Studies were performed to evaluate potential cross-contamination on the **cobas**® 6800/8800 Systems using **cobas**® CT/NG. Cross-contamination can cause false positive results. In this performance study the sample-to-sample cross-contamination rate of **cobas**® CT/NG has been determined to be 0.5% (2/432), (95% CI: 0.1%-1.7%) when alternating very high positive and negative samples were tested over nine runs. Run-to-run cross-contamination has not been observed (0/282). Testing was done using samples prepared with **cobas**® PCR Media and with PreservCyt® Solution and with urine stabilized in **cobas**® PCR Media. High positive samples ($\geq 10^3$ IFU/mL for CT and $\geq 10^4$ CFU/mL for NG) in the study were prepared to generate a Ct value that was lower than that obtained with 95% or more of the specimens of infected patients in

the intended use population. Cross contamination rates in clinical settings depend on the proportion of high positive samples and prevalence of the disease. Routine clinical cross-contamination rates are expected to be lower than what was observed in this study and need to be assessed in user's settings.

Clinical performance evaluation

The clinical utility and performance of cobas® CT/NG was established in a multi-site, prospective collection study by comparing the results to a Patient Infected Status (PIS) that used a combination of FDA-cleared NAATs for urogenital specimens. Female and male urogenital specimens were collected at 9 geographically diverse sites in the US with testing performed at 4 laboratory testing sites (3 external and 1 internal).

Prospectively enrolled female subjects provided the following urogenital specimens: first-void urine, 3 vaginal swabs, 1 endocervical swab in **cobas**® PCR Media, and 1 cervical sample in PreservCyt® Solution. If the female was in the clinician-collected vaginal swab arm of the study, 2 of the vaginal swabs were placed in the respective manufacturers' collection device and 1 in **cobas**® PCR Media. If the female subject was in the self-collected vaginal swab self-collection arm of the study, then 1 vaginal swab was self-collected first and placed into **cobas**® PCR Media and then followed by the 2 clinician-collected vaginal swabs and placed in the 2 respective manufacturers' collection devices.

Prospectively enrolled male subjects provided a urine specimen that was aliquoted into the respective manufacturers' collection device and **cobas**® PCR Media.

Subjects were classified as symptomatic if they self-reported symptoms indicative of a CT or NG infection as listed below:

- Dysuria (pain during urination)
- Coital pain, difficulty or bleeding
- Pelvic pain
- Abnormal vaginal discharge
- Pelvic, uterine or ovarian pain
- Urethral discharge
- Testicular pain
- Scrotal pain or swelling

Prospectively enrolled subjects were classified as asymptomatic if they did not report any of the above symptoms.

Specimens were tested for CT and NG using **cobas**® CT/NG and commercially available NAATs. All tests were run according to the respective manufacturers' Instructions For Use.

The clinical performance of **cobas**® CT/NG was evaluated by comparing the results from collected specimen types to a pre-specified PIS (Patient Infected Status) algorithm as determined by the combined results from 2 commercially available NAATs for females and 3 commercially available NAATs for males. The PIS algorithms for Female and Male subjects are shown in Table 24 and Table 25, respectively.

For NG, archived prospectively collected female urine, cervical specimens in PreservCyt®, and endocervical swabs were obtained from the clinical study for **cobas**® CT/NG v2 test on the **cobas**® 4800 System. The PIS of these specimens were already determined from the clinical study for **cobas**® CT/NG v2 test on the **cobas**® 4800 System.

Table 24 Determination of female Patient Infected Status (PIS) for urogenital specimens[a]

NAAT1 Urine/Vaginal	NAAT2 Urine/Vaginal	Patient Infected Status (PIS)[b]
+/+	+/+	Infected
+/+	+/- or -/+	Infected
+/- or -/+	+/+	Infected
+/-	-/+	Infected
-/+	+/- or -/+	Infected
+/-	+/-	Infected (Urine) Non-Infected (Vaginal)
+/- or -/+	-/-	Not Infected
+/+	-/-	Not Infected
-/-	+/+	Not Infected
-/-	+/- or -/+	Not Infected
-/-	-/-	Not Infected

[a] One or more positives in each NAAT (NAAT1 and NAAT2) designates the PIS as positive. Any other combination of results defines the PIS as negative.

[b] In the scenario where one or more of the sample types are invalid, the remaining sample types with valid results from NAAT1 and NAAT2 must have concordant positive or concordant negative results to determine the PIS as Infected or Not Infected, respectively. For all other cases where one or more of the sample types are invalid, the PIS is indeterminate.

Table 25 Determination of male Patient Infected Status (PIS) for urine specimens

NAAT1 Urine	NAAT2 Urine	NAAT3 Urine	Patient Infected Status (PIS)[a]
+	+	+	Infected
+	+	-	Infected
+	-	+	Infected
-	+	+	Infected
-	-	+	Not Infected
-	+	-	Not Infected
+	-	-	Not Infected
-	-	-	Not Infected

[a] If at least 2 out of the 3 test results are concordant positive or negative then the PIS can be considered as infected or non-infected, respectively. If one test result is invalid/missing and the other two test results are discordant then the PIS is indeterminate. If 2 or 3 test results are invalid/missing, then the PIS is indeterminate.

Sensitivity (SENS), specificity (SPEC), positive predictive value (PPV), and negative predictive value (NPV) of **cobas®** CT/NG were calculated separately for the detection of CT or NG using the PIS as the composite reference standard and evaluated by gender, sample type, and symptom status. In addition, the predictive values were calculated based on overall sensitivity and specificity (with all data combined for males and females) for a range of hypothetical prevalence values.

Results

A total of 5,197 subjects were prospectively enrolled, of which 5,105 were eligible for inclusion. Of the 5,105 eligible subjects contributing prospective specimens, 5,053 (99.0%) (3,860 females and 1,193 males) were evaluable and were included in the data analyses. A total of 52 subjects (1.0%) were classified as non-evaluable and excluded from all statistical analyses. There were a total of 371 archived prospectively collected female urogenital samples (urine, cervical specimens in PreservCyt, and endocervical swabs) tested in this clinical study from 295 female subjects. Among the 17,169 samples tested in this study, 19 samples exhibited invalid results on the first run (invalid rate of 0.11% (95%CI: 0.07% ; 0.17%)). Upon repeat testing, 3 samples exhibited valid results.

Table 26 and Table 27 summarize the results from symptomatic and asymptomatic, prospectively enrolled subjects designated as infected or non-infected with CT (females and males, respectively) according to the PIS algorithm. A total of 271 females and 118 males were infected with CT. Symptoms were reported in 45.8% (124/271) of infected and 36.7% (1318/3589) of non-infected females. Symptoms were reported in 53.4% (63/118) of infected and 22.5% (242/1074) of non-infected males.

Table 26 CT positive/negative analyses for female Patient Infected Status

	NAAT1		NAAT2		cobas® CT/NG				Symptom Status[a]		
Patient Infected Status	**UR**	**VS**	**UR**	**VS**	**UR**	**VS**	**PC**	**ES**	**Symp**	**Asymp**	**Total**
Infected	+	+	+	+	+	+	+	+	104	108	212
Infected	-	+	+	+	+	+	+	+	2	7	9
Infected	+	+	+	+	+	+	-	+	1	5	6
Infected	+	+	-	+	+	+	+	+	2	4	6
Infected	+	+	+	+	+	+	-	-	1	4	5
Infected	+	+	+	+	+	+	+	-	1	3	4
Infected	-	+	+	+	-	+	+	+	1	3	4
Infected	-	+	-	+	-	+	+	+	2	2	4
Infected	-	+	-	+	+	+	+	+	2	1	3
Infected	+	-	+	+	+	+	-	-	1	1	2
Infected	+	+	+	+	+	+	Failed	+	0	1	1
Infected	+	+	+	+	+	+	+	Failed	1	0	1
Infected	-	+	+	+	+	-	+	+	0	1	1
Infected	-	+	+	+	+	+	+	-	0	1	1
Infected	-	+	+	+	+	+	-	+	1	0	1
Infected	-	+	+	+	+	+	-	-	0	1	1
Infected	-	+	+	+	-	+	-	-	1	0	1
Infected	-	+	-	+	+	+	+	-	0	1	1
Infected	-	+	-	+	-	+	-	+	0	1	1

	NAAT1		NAAT2		cobas® CT/NG				Symptom Status[a]		
Patient Infected Status	**UR**	**VS**	**UR**	**VS**	**UR**	**VS**	**PC**	**ES**	**Symp**	**Asymp**	**Total**
Infected	+	-	+	+	+	+	+	+	1	0	1
Infected	+	-	+	+	+	-	-	-	0	1	1
Infected	+	-	-	+	-	+	-	-	1	0	1
Infected	+	-	-	+	-	-	-	-	0	1	1
Infected[b]	+	-	+	-	+	+	-	+	1	0	1
Infected[b]	+	-	+	-	+	+	-	-	1	0	1
Infected[b]	+	-	+	-	+	-	-	-	0	1	1
Total									124	147	271
Non-Infected	-	-	-	-	-	-	-	-	1252	2165	3417
Non-Infected	-	-	-	+	-	-	-	-	6	12	18
Non-Infected	-	Invalid	-	-	-	-	-	-	7	5	12
Non-Infected	-	-	-	-	-	Invalid	-	-	6	4	10
Non-Infected	-	-	-	-	-	+	-	-	1	9	10
Non-Infected	-	-	+	-	-	-	-	-	2	7	9
Non-Infected	-	-	-	-	-	-	-	+	5	4	9
Non-Infected	-	-	NA	-	-	-	-	-	2	7	9
Non-Infected	-	-	Invalid	-	-	-	-	-	0	9	9
Non-Infected	-	-	-	-	-	-	-	NA	3	5	8
Non-Infected	+	-	-	-	-	-	-	-	3	3	6
Non-Infected	-	-	-	-	+	-	-	-	1	5	6
Non-Infected	-	+	-	-	-	-	-	-	2	2	4
Non-Infected	-	-	-	-	-	-	+	-	1	3	4
Non-Infected	-	-	-	-	-	NA	-	-	1	3	4
Non-Infected	-	NA	-	-	-	-	-	-	0	4	4
Non-Infected	-	-	-	-	-	-	NA	NA	0	3	3
Non-Infected	-	-	-	NA	-	-	-	-	1	2	3
Non-Infected	NA	-	-	-	-	-	-	-	0	3	3
Non-Infected	Invalid	-	-	-	-	-	-	-	2	1	3
Non-Infected	-	+	-	-	-	+	-	-	2	0	2
Non-Infected	-	-	-	+	-	+	+	+	0	2	2
Non-Infected	-	-	-	+	-	+	+	-	2	0	2
Non-Infected	-	-	-	+	-	+	-	+	1	1	2
Non-Infected	-	-	-	+	-	+	-	-	0	2	2
Non-Infected	-	-	-	-	-	+	+	+	2	0	2

	NAAT1		**NAAT2**		**cobas® CT/NG**				**Symptom Status[a]**		
Patient Infected Status	**UR**	**VS**	**UR**	**VS**	**UR**	**VS**	**PC**	**ES**	**Symp**	**Asymp**	**Total**
Non-Infected	-	-	-	-	-	-	-	Invalid	2	0	2
Non-Infected	-	-	-	Invalid	-	-	-	-	1	1	2
Non-Infected	-	-	+	+	-	+	-	-	0	1	1
Non-Infected	-	-	+	-	+	-	-	-	0	1	1
Non-Infected	-	-	+	-	-	+	-	-	1	0	1
Non-Infected	-	-	-	+	+	+	+	+	0	1	1
Non-Infected	-	-	-	+	-	-	+	+	1	0	1
Non-Infected	-	-	-	+	+	+	-	+	0	1	1
Non-Infected	-	-	-	+	-	-	-	+	0	1	1
Non-Infected	-	-	-	+	+	+	-	-	0	1	1
Non-Infected	-	-	-	-	-	-	NA	-	0	1	1
Non-Infected	-	-	-	-	-	Invalid	Invalid	Invalid	1	0	1
Non-Infected	-	-	-	-	-	-	Invalid	Invalid	1	0	1
Non-Infected	-	-	-	-	-	NA	Invalid	-	1	0	1
Non-Infected	-	-	-	-	-	-	Invalid	-	1	0	1
Non-Infected	-	-	-	-	-	+	+	-	1	0	1
Non-Infected	-	-	-	-	-	-	-	Failed	1	0	1
Non-Infected	-	-	-	-	+	-	-	+	1	0	1
Non-Infected	-	-	-	-	-	+	-	+	1	0	1
Non-Infected	-	-	-	-	Failed	-	-	-	1	0	1
Non-Infected	-	-	-	-	-	Failed	-	-	1	0	1
Non-Infected	-	-	-	Invalid	-	+	+	+	0	1	1
Non-Infected	-	-	NA	+	-	+	-	-	0	1	1
Non-Infected	-	Invalid	-	-	-	Invalid	-	-	1	0	1
Total									1318	2271	3589

	NAAT1		NAAT2		cobas® CT/NG				Symptom Status[a]		
Patient Infected Status	**UR**	**VS**	**UR**	**VS**	**UR**	**VS**	**PC**	**ES**	**Symp**	**Asymp**	**Total**

[a] Symp = symptomatic, Asymp = asymptomatic.
[b] Infected (Urine), Non-Infected (Swabs).

Note: In the scenario where one or more of the sample types are invalid/not available (NA), for female subjects, the remaining sample types with valid results from NAAT1 and NAAT2 must have concordant positive or concordant negative results to determine the PIS as Infected or Not Infected, respectively. For all other cases where one or more of the sample types are invalid/not available (NA), the PIS is indeterminate.

Note: Female subjects with designated infection status (Infected or Non-Infected) and final valid **cobas®** CT/NG test results are considered evaluable and included in this summary table.
Note: + denotes Positive, - denotes Negative, NA denotes Not Available.
Note: UR = urine, VS = vaginal swab, PC = PreservCyt®, ES = endocervical swab.

Note: **cobas®** Invalid are the sum of instrument amplification/detection errors and samples excluded due to protocol deviations

Note: **cobas®** Failed are hardware, software or operator errors causing no result reported

Table 27 CT positive/negative analysis for male Patient Infected Status

	NAAT1	**NAAT2**	**NAAT3**	**cobas® CT/NG**	**Symptom Status[a]**		
Patient Infected Status	**UR**	**UR**	**UR**	**UR**	**Symp**	**Asymp**	**Total**
Infected	+	+	+	+	60	55	115
Infected	-	+	+	+	1	0	1
Infected	+	Invalid	+	+	1	0	1
Infected	+	-	+	+	1	0	1
Total Infected					63	55	118
Non-Infected	-	-	-	-	238	819	1057
Non-Infected	-	Invalid	-	-	2	2	4
Non-Infected	Invalid	-	-	-	0	3	3
Non-Infected	-	-	Invalid	-	0	3	3
Non-Infected	NA	-	-	-	1	1	2
Non-Infected	-	-	-	+	0	2	2
Non-Infected	-	-	+	-	0	1	1
Non-Infected	-	+	-	+	1	0	1
Non-Infected	+	-	-	-	0	1	1
Total Non-Infected					242	832	1074*

[a] Symp = symptomatic, Asymp = asymptomatic.

*One subject was missing symptom status and is not presented in this table.

Note: If at least 2 out of the 3 test results, for male subjects, are concordant positive or negative then the PIS can be considered as infected or non-infected, respectively. If one test result is invalid/not available (NA) and the other two test results are discordant then the PIS is indeterminate. If 2 or 3 test results are invalid/not available, then the PIS is indeterminate.

Note: Male subjects with designated patient infection status (Infected or Non-Infected) and final valid **cobas®** CT/NG test results are considered evaluable and included in this summary table.

Note: **cobas®** Invalid are the sum of instrument amplification/detection errors and samples excluded due to protocol deviations
Note: + denotes Positive, - denotes Negative, NA denotes Not Available.
Note: UR = urine.

Sensitivity, specificity, and predictive values of **cobas®** CT/NG for CT as defined by PIS are presented by gender, sample type, and symptom status in Table 28.

Table 28 CT clinical performance compared with Patient Infected Status by gender, sample type, and symptom status

Sample Type[a]	Symptom Status[b]	Total (n)	SENS	95% Score CI	SPEC	95% Score CI	PREV (%)	PPV (%)	NPV (%)
Female									
UR	Symp	1441	96.0% (119/124)	(90.9%, 98.3%)	99.8% (1315/1317)	(99.4%, 100.0%)	8.6	98.3	99.6
	Asymp	2418	95.2% (140/147)	(90.5%, 97.7%)	99.6% (2262/2271)	(99.2%, 99.8%)	6.1	94.0	99.7
	Overall	3859	95.6% (259/271)[c]	(92.4%, 97.4%)	99.7% (3577/3588)	(99.5%, 99.8%)	7.0	95.9	99.7
VS-C	Symp	711	100.0% (63/63)	(94.3%, 100.0%)	99.2% (643/648)	(98.2%, 99.7%)	8.9	92.6	100.0
	Asymp	1225	97.6% (83/85)	(91.8%, 99.4%)	99.0% (1129/1140)	(98.3%, 99.5%)	6.9	88.3	99.8
	Overall	1936	98.6% (146/148)	(95.2%, 99.6%)	99.1% (1772/1788)	(98.6%, 99.4%)	7.6	90.1	99.9
VS-S	Symp	720	100.0% (59/59)	(93.9%, 100.0%)	98.8% (653/661)	(97.6%, 99.4%)	8.2	88.1	100.0
	Asymp	1186	98.4% (60/61)	(91.3%, 99.7%)	99.2% (1116/1125)	(98.5%, 99.6%)	5.1	87.0	99.9
	Overall	1906	99.2% (119/120)	(95.4%, 99.9%)	99.0% (1769/1786)	(98.5%, 99.4%)	6.3	87.5	99.9
PC	Symp	1438	95.1% (116/122)	(89.7%, 97.7%)	99.5% (1309/1316)	(98.9%, 99.7%)	8.5	94.3	99.5
	Asymp	2413	90.3% (131/145)	(84.4%, 94.2%)	99.7% (2261/2268)	(99.4%, 99.9%)	6.0	94.9	99.4
	Overall	3851	92.5% (247/267)	(88.7%, 95.1%)	99.6% (3570/3584)	(99.3%, 99.8%)	6.9	94.6	99.4
ES	Symp	1433	95.9% (116/121)	(90.7%, 98.2%)	99.1% (1300/1312)	(98.4%, 99.5%)	8.4	90.6	99.6
	Asymp	2410	91.1% (133/146)	(85.4%, 94.7%)	99.5% (2253/2264)	(99.1%, 99.7%)	6.1	92.4	99.4
	Overall	3843	93.3% (249/267)	(89.6%, 95.7%)	99.4% (3553/3576)	(99.0%, 99.6%)	6.9	91.5	99.5

Sample Type[a]	Symptom Status[b]	Total (n)	SENS	95% Score CI	SPEC	95% Score CI	PREV (%)	PPV (%)	NPV (%)
Male									
UR	Symp	305	100.0% (63/63)	(94.3%, 100.0%)	99.6% (241/242)	(97.7%, 99.9%)	20.7	98.4	100.0
	Asymp	887	100.0% (55/55)	(93.5%, 100.0%)	99.8% (830/832)	(99.1%, 99.9%)	6.2	96.5	100.0
	Overall	1192*	100.0% (118/118)	(96.8%, 100.0%)	99.7% (1071/1074)	(99.2%, 99.9%)	9.9	97.5	100.0

[a] UR = urine, VS-C = clinician-collected vaginal swab, VS-S = self-collected vaginal swab, PC = PreservCyt®, ES = endocervical swab.

[b] Symp = symptomatic, Asymp = asymptomatic.

[c] Five CT PIS infected females had a CT negative urine specimen with NAAT1 and NAAT2 while they had a CT positive vaginal swab with NAAT1 and NAAT2.

* One subject was missing symptom status and is not presented in this table.

Note: In the scenario where one or more of the sample types are invalid/not available, for female subjects, the remaining sample types with valid results from NAAT1 and NAAT2 must have concordant positive or concordant negative results to determine the PIS as Infected or Non-Infected, respectively. For all other cases where one or more of the sample types are invalid/not available, the PIS is indeterminate.

Note: If at least 2 out of the 3 test results, for male subjects, are concordant positive or negative then the PIS can be considered as infected or non-infected, respectively. If one test result is invalid/not available and the other two test results are discordant then the PIS is indeterminate. If 2 or 3 test results are invalid/not available, then the PIS is indeterminate.

Note: Subjects with designated patient infection status (Infected or Non-Infected) and final valid **cobas**® CT/NG test results are considered evaluable and included in this summary table. An evaluable subject may not have all available sample types or valid test results.

Note: CI = confidence interval, PREV = prevalence, SENS = sensitivity, SPEC = specificity, PPV = positive predictive value, NPV = negative predictive value.

Table 29 and Table 30 summarize the results from symptomatic and asymptomatic subjects designated as infected or non-infected with NG (females and males, respectively) according to the PIS algorithm. A total of 57 females and 87 males were infected with NG. Symptoms were reported in 45.6% (26/57) of infected and 37.2% (1416/3803) of non-infected females. Symptoms were reported in 94.3% (82/87) of infected and 20.2% (223/1105) of non-infected males.

Table 29 NG positive/negative analysis for female Patient Infected Status (prospective specimens)

	NAAT1		**NAAT2**		**cobas® CT/NG**				**Symptom Status[a]**		
Patient Infected Status	**UR**	**VS**	**UR**	**VS**	**UR**	**VS**	**PC**	**ES**	**Symp**	**Asymp**	**Total**
Infected	+	+	+	+	+	+	+	+	20	23	43
Infected	-	+	-	+	-	+	+	+	2	3	5
Infected	+	+	-	+	+	+	+	+	0	2	2
Infected	-	+	-	+	+	+	+	+	2	0	2
Infected	+	+	+	+	+	+	+	Failed	1	0	1
Infected	+	+	+	+	+	+	-	-	0	1	1
Infected	+	+	-	+	-	+	-	-	0	1	1
Infected	-	+	NA	+	+	+	+	-	0	1	1
Infected[b]	+	-	+	-	+	-	-	-	1	0	1
Total									26	31	57
Non-Infected	-	-	-	-	-	-	-	-	1368	2315	3683
Non-Infected	-	+	-	-	-	-	-	-	4	11	15
Non-Infected	+	-	-	-	-	-	-	-	5	7	12
Non-Infected	-	-	NA	-	-	-	-	-	2	7	9
Non-Infected	-	Invalid	-	-	-	-	-	-	5	4	9
Non-Infected	-	-	-	-	-	Invalid	-	-	5	3	8
Non-Infected	-	-	-	-	-	-	-	NA	3	5	8
Non-Infected	-	-	Invalid	-	-	-	-	-	0	8	8
Non-Infected	-	-	-	-	-	+	-	-	2	4	6
Non-Infected	-	-	-	-	-	NA	-	-	1	3	4
Non-Infected	-	NA	-	-	-	-	-	-	0	4	4
Non-Infected	-	-	-	-	-	-	NA	NA	0	3	3
Non-Infected	-	-	-	NA	-	-	-	-	1	2	3
Non-Infected	NA	-	-	-	-	-	-	-	0	3	3
Non-Infected	Invalid	-	-	-	-	-	-	-	2	1	3
Non-Infected	+	+	-	-	-	+	-	-	0	2	2
Non-Infected	-	-	-	-	+	-	-	-	2	0	2
Non-Infected	-	-	-	-	-	-	-	Invalid	2	0	2
Non-Infected	+	+	-	-	-	-	-	-	0	1	1
Non-Infected	+	+	-	-	-	Invalid	-	-	1	0	1
Non-Infected	-	+	-	-	-	+	+	-	1	0	1
Non-Infected	-	+	-	-	-	+	-	-	1	0	1

	NAAT1		NAAT2		cobas® CT/NG				Symptom Status[a]		
Patient Infected Status	**UR**	**VS**	**UR**	**VS**	**UR**	**VS**	**PC**	**ES**	**Symp**	**Asymp**	**Total**
Non-Infected	-	-	+	+	+	-	-	-	1	0	1
Non-Infected	-	-	-	+	-	+	-	-	0	1	1
Non-Infected	-	-	-	-	-	-	+	+	0	1	1
Non-Infected	-	-	-	-	-	-	-	+	1	0	1
Non-Infected	-	-	-	-	-	Failed	-	-	1	0	1
Non-Infected	-	-	-	-	Failed	-	-	-	1	0	1
Non-Infected	-	-	-	-	-	-	-	Failed	1	0	1
Non-Infected	-	-	-	-	-	-	NA	-	0	1	1
Non-Infected	-	-	-	-	-	-	Invalid	-	1	0	1
Non-Infected	-	-	-	-	-	NA	Invalid	-	1	0	1
Non-Infected	-	-	-	-	-	-	Invalid	Invalid	1	0	1
Non-Infected	-	-	-	-	-	Invalid	Invalid	Invalid	1	0	1
Non-Infected	-	-	-	-	-	-	Failed	-	0	1	1
Non-Infected	-	-	-	Invalid	-	-	-	-	1	0	1
Non-Infected	-	Invalid	-	-	-	Invalid	-	-	1	0	1
Total									1416	2387	3803

[a] Symp = symptomatic, Asymp = asymptomatic.
[b] Infected (Urine), Non-Infected (Swabs).

Note: In the scenario where one or more of the sample types are invalid/not available (NA), for female subjects, the remaining sample types with valid results from NAAT1 and NAAT2 must have concordant positive or concordant negative results to determine the PIS as Infected or Not Infected, respectively. For all other cases where one or more of the sample types are invalid/not available (NA), the PIS is indeterminate.

Note: Female subjects with designated infection status (Infected or Non-Infected) and final valid **cobas®** CT/NG test results are considered evaluable and included in this summary table.

Note: + denotes Positive, - denotes Negative, NA denotes Not Available.
Note: UR = urine, VS = vaginal swab, PC = PreservCyt®, ES = endocervical swab.

Note: **cobas®** Invalid are the sum of instrument amplification/detection errors and samples excluded due to protocol deviations

Note: **cobas®** Failed are hardware, software or operator errors causing no result reported

Table 30 NG positive/negative analysis for male Patient Infected Status

	NAAT1	**NAAT2**	**NAAT3[a]**	**cobas® CT/NG**	**Symptom Status[a]**		
Patient Infected Status	**UR**	**UR**	**UR**	**UR**	**Symp**	**Asymp**	**Total**
Infected	+	+	+	+	81	5	86
Infected	NA	+	+	+	1	0	1
Total Infected					82	5	87
Non-Infected	-	-	-	-	215	863	1078
Non-Infected	+	-	-	-	2	7	9
Non-Infected	-	Invalid	-	-	3	2	5
Non-Infected	-	-	-	+	2	2	4
Non-Infected	Invalid	-	-	-	0	3	3
Non-Infected	-	-	Invalid	-	0	3	3
Non-Infected	-	+	-	+	1	1	2
Non-Infected	NA	-	-	-	0	1	1
Total Non-Infected					223	882	1105*

[a] Symp = symptomatic, Asymp = asymptomatic.

*One subject was missing symptom status and is not included in this table.

Note: If at least 2 out of the 3 test results, for male subjects, are concordant positive or negative then the PIS can be considered as Infected or Non-Infected, respectively. If one test result is invalid/not available (NA) and the other two test results are discordant then the PIS is indeterminate. If 2 or 3 test results are invalid/not available, then the PIS is indeterminate.

Note: Male subjects with designated patient infection status (Infected or Non-Infected) and final valid **cobas®** CT/NG test results are considered evaluable and included in this summary table.

Note: **cobas®** Invalid are the sum of instrument amplification/detection errors and samples excluded due to protocol deviations

Note: + denotes Positive, - denotes Negative, NA denotes Not Available.

Note: UR = urine.

Sensitivity, specificity, and predictive values of **cobas®** CT/NG for NG as defined by PIS are presented by gender, sample type, and symptom status in Table 31 (prospective and archived prospectively collected specimens).

Table 31 NG clinical performance compared with Patient Infected Status by gender, sample type, and symptom status (prospective and archived prospectively collected specimens)

Sample Type[a]	Symptom Status[b]	Total (n)	SENS	95% Score CI	SPEC	95% Score CI	PREV (%)	PPV (%)	NPV (%)
Female									
UR (prospective)	Symp	1441	92.3% (24/26)	(75.9%, 97.9%)	99.8% (1412/1415)	(99.4%, 99.9%)	1.8	88.9	99.9
	Asymp	2418	87.1% (27/31)	(71.1%, 94.9%)	100.0% (2387/2387)	(99.8%, 100.0%)	1.3	100.0	99.8
	Overall	3859	89.5% (51/57) [c]	(78.9%, 95.1%)	99.9% (3799/3802)	(99.8%, 100.0%)	1.5	94.4	99.8
UR (archived)	Symp	94	100.0% (35/35)	(90.1%, 100.0%)	100.0% (59/59)	(93.9%, 100.0%)	37.2	100.0	100.0
	Asymp	101	97.6% (41/42)	(87.7%, 99.6%)	100.0% (59/59)	(93.9%, 100.0%)	41.6	100.0	98.3
	Overall	195	98.7% (76/77)	(93.0%, 99.8%)	100.0% (118/118)	(96.8%, 100.0%)	39.5	100.0	99.2
UR (prospective and archived)	Symp	1535	96.7% (59/61)	(88.8%, 99.1%)	99.8% (1471/1474)	(99.4%, 99.9%)	4.0	95.2	99.9
	Asymp	2519	93.2% (68/73)	(84.9%, 97.0%)	100.0% (2446/2446)	(99.8%, 100.0%)	2.9	100.0	99.8
	Overall	4054	94.8% (127/134)	(89.6%, 97.4%)	99.9% (3917/3920)	(99.8%, 100.0%)	3.3	97.7	99.8
VS-C	Symp	711	100.0% (11/11)	(74.1%, 100.0%)	99.7% (698/700)	(99.0%, 99.9%)	1.5	84.6	100.0
	Asymp	1225	100.0% (17/17)	(81.6%, 100.0%)	99.8% (1205/1208)	(99.3%, 99.9%)	1.4	85.0	100.0
	Overall	1936	100.0% (28/28)	(87.9%, 100.0%)	99.7% (1903/1908)	(99.4%, 99.9%)	1.4	84.8	100.0
VS-S	Symp	720	100.0% (14/14)	(78.5%, 100.0%)	99.7% (704/706)	(99.0%, 99.9%)	1.9	87.5	100.0
	Asymp	1187	100.0% (14/14)	(78.5%, 100.0%)	99.7% (1169/1173)	(99.1%, 99.9%)	1.2	77.8	100.0
	Overall	1907	100.0% (28/28)	(87.9%, 100.0%)	99.7% (1873/1879)	(99.3%, 99.9%)	1.5	82.4	100.0
PC (prospective)	Symp	1438	100.0% (25/25)	(86.7%, 100.0%)	99.9% (1412/1413)	(99.6%, 100.0%)	1.7	96.2	100.0
	Asymp	2413	93.5% (29/31)	(79.3%, 98.2%)	100.0% (2381/2382)	(99.8%, 100.0%)	1.3	96.7	99.9
	Overall	3851	96.4% (54/56)	(87.9%, 99.0%)	99.9% (3793/3795)	(99.8%, 100.0%)	1.5	96.4	99.9

Sample Type[a]	Symptom Status[b]	Total (n)	SENS	95% Score CI	SPEC	95% Score CI	PREV (%)	PPV (%)	NPV (%)
Female									
PC (archived)	Symp	48	95.7% (22/23)	(79.0%, 99.2%)	100.0% (25/25)	(86.7%, 100.0%)	47.9	100.0	96.2
	Asymp	23	100.0% (10/10)	(72.2%, 100.0%)	100.0% (13/13)	(77.2%, 100.0%)	43.5	100.0	100.0
	Overall	71	97.0% (32/33)	(84.7%, 99.5%)	100.0% (38/38)	(90.8%, 100.0%)	46.5	100.0	97.4
PC (prospective and archived)	Symp	1486	97.9% (47/48)	(89.1%, 99.6%)	99.9% (1437/1438)	(99.6%, 100.0%)	3.2	97.9	99.9
	Asymp	2436	95.1% (39/41)	(83.9%, 98.7%)	100.0% (2394/2395)	(99.8%, 100.0%)	1.7	97.5	99.9
	Overall	3922	96.6% (86/89)	(90.6%, 98.8%)	99.9% (3831/3833)	(99.8%, 100.0%)	2.3	97.7	99.9
ES (prospective)	Symp	1433	100.0% (24/24)	(86.2%, 100.0%)	99.9% (1408/1409)	(99.6%, 100.0%)	1.7	96.0	100.0
	Asymp	2410	90.3% (28/31)	(75.1%, 96.7%)	100.0% (2378/2379)	(99.8%, 100.0%)	1.3	96.6	99.9
	Overall	3843	94.5% (52/55)	(85.1%, 98.1%)	99.9% (3786/3788)	(99.8%, 100.0%)	1.4	96.3	99.9
ES (archived)	Symp	51	100.0% (21/21)	(84.5%, 100.0%)	100.0% (30/30)	(88.6%, 100.0%)	41.2	100.0	100.0
	Asymp	54	100.0% (24/24)	(86.2%, 100.0%)	100.0% (30/30)	(88.6%, 100.0%)	44.4	100.0	100.0
	Overall	105	100.0% (45/45)	(92.1%, 100.0%)	100.0% (60/60)	(94.0%, 100.0%)	42.9	100.0	100.0
ES (prospective and archived)	Symp	1484	100.0% (45/45)	(92.1%, 100.0%)	99.9% (1438/1439)	(99.6%, 100.0%)	3.0	97.8	100.0
	Asymp	2464	94.5% (52/55)	(85.1%, 98.1%)	100.0% (2408/2409)	(99.8%, 100.0%)	2.2	98.1	99.9
	Overall	3948	97.0% (97/100)	(91.5%, 99.0%)	99.9% (3846/3848)	(99.8%, 100.0%)	2.5	98.0	99.9

Sample Type[a]	Symptom Status[b]	Total (n)	SENS	95% Score CI	SPEC	95% Score CI	PREV (%)	PPV (%)	NPV (%)
Male									
UR	Symp	305	100.0% (82/82)	(95.5%, 100.0%)	98.7% (220/223)	(96.1%, 99.5%)	26.9	96.5	100.0
	Asymp	887	100.0% (5/5)	(56.6%, 100.0%)	99.7% (879/882)	(99.0%, 99.9%)	0.6	62.5	100.0
	Overall	1192*	100.0% (87/87)	(95.8%, 100.0%)	99.5% (1099/1105)	(98.8%, 99.8%)	7.3	93.5	100.0

[a] UR = urine, VS-C = clinician-collected vaginal swab, VS-S = self-collected vaginal swab, PC = PreservCyt®, ES = endocervical swab.
[b] Symp = symptomatic, Asymp = asymptomatic.
[c] Five NG PIS infected females had a NG negative urine specimen with NAAT1 and NAAT2 while they had a NG positive vaginal swab with NAAT1 and NAAT2.
* One subject was missing symptom status and is not included in this table.

Note: In the scenario where one or more of the sample types are invalid/not available, for female subjects, the remaining sample types with valid results from NAAT1 and NAAT2 must have concordant positive or concordant negative results to determine the PIS as Infected or Non-Infected, respectively. For all other cases where one or more of the sample types are invalid/ not available, the PIS is indeterminate.

Note: If at least 2 out of the 3 test results, for male subjects, are concordant positive or negative then the PIS can be considered as Infected or Non-Infected, respectively. If one test result is invalid/not available and the other two test results are discordant then the PIS is indeterminate. If 2 or 3 test results are invalid/not available, then the PIS is indeterminate.

Note: Subjects with designated patient infection status (Infected or Non-Infected) and final valid **cobas**® CT/NG test results are considered evaluable and included in this summary table. An evaluable subject may not have all available sample types or valid test results.

Note: Archived prospectively collected specimens were from COB-CTNG-282 study and included female PIS positive subjects that have available sample with adequate volume for testing.

Note: CI = confidence interval, PREV = prevalence, SENS = sensitivity, SPEC = specificity, PPV = positive predictive value, NPV = negative predictive value.

Expected values

Prevalence

The prevalence of CT and NG in patient populations depends on a variety of factors including age, gender, the presence of symptoms, clinic type, and test method. The positivity rate of CT observed with **cobas**® CT/NG during this multi-site clinical study was 7.2% overall. The overall positivity rate of NG observed with **cobas**® CT/NG for the prospective and archived prospectively collected samples was 2.7% .

Positive and negative predictive values

The positive and negative predictive values of all *in vitro* diagnostic tests are highly dependent on prevalence. The **cobas**® CT/NG performance may vary depending on the prevalence and the population tested. Hypothetical positive and negative values (PPV and NPV) derived from disease prevalence of 1 to 50% for **cobas**® CT/NG are shown in Table 32 and Table 33. These tables use the overall sensitivity and specificity (compared with PIS) across all sample types in both female and male subjects: 95.5% and 99.5% respectively for CT, and 96.5% and 99.9% respectively for NG.

Table 32 Positive and negative predictive values for hypothetical CT prevalence

Prevalence (%)	Sensitivity [a] (%)	Specificity [a] (%)	PPV (%)	NPV (%)
1	95.5	99.5	63.89	99.95
3	95.5	99.5	84.41	99.86
5	95.5	99.5	90.21	99.77
10	95.5	99.5	95.11	99.51
15	95.5	99.5	96.87	99.22
20	95.5	99.5	97.77	98.89
30	95.5	99.5	98.69	98.12
50	95.5	99.5	99.43	95.72

Note: PPV = Positive predictive value, NPV = Negative predictive value.

[a] The overall sensitivity and specificity were estimated by comparing the **cobas**® CT/NG Test results to patient infected status across all sample types in both female and male subjects.

Table 33 Positive and negative predictive values for hypothetical NG prevalence (Prospective specimen only)

Prevalence (%)	Sensitivity [a] (%)	Specificity [a] (%)	PPV (%)	NPV (%)
1	96.5	99.9	86.86	99.96
3	96.5	99.9	95.29	99.89
5	96.5	99.9	97.18	99.81
10	96.5	99.9	98.64	99.61
15	96.5	99.9	99.14	99.38
20	96.5	99.9	99.39	99.12
30	96.5	99.9	99.64	98.50
50	96.5	99.9	99.85	96.58

Note: PPV = Positive predictive value, NPV = Negative predictive value.

[a] The overall sensitivity and specificity were estimated by comparing the **cobas**® CT/NG Test results to patient infected status across all sample types in both female and male subjects.

Cycle threshold frequency distribution

A total of 1237 specimens (female and male) were positive for CT and a total of 481 specimens (female and male) were

positive for NG. The frequency distribution of **cobas**® CT/NG positive results for CT and NG infected specimens are shown in Figure 5 and Figure 6, respectively.

Figure 5 Cycle threshold distribution of CT positive specimens

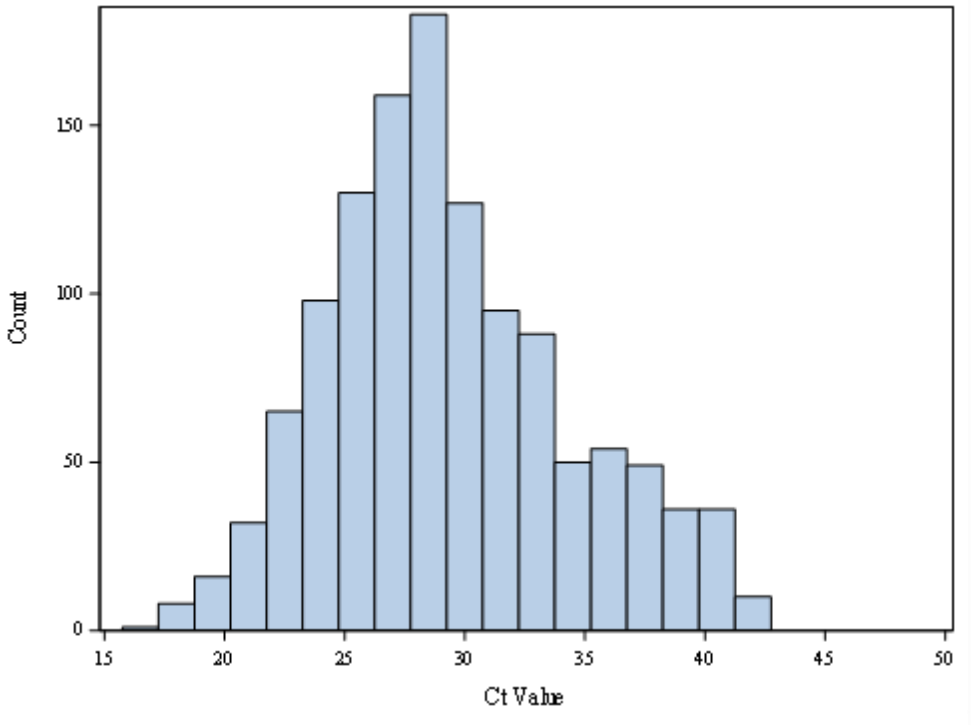
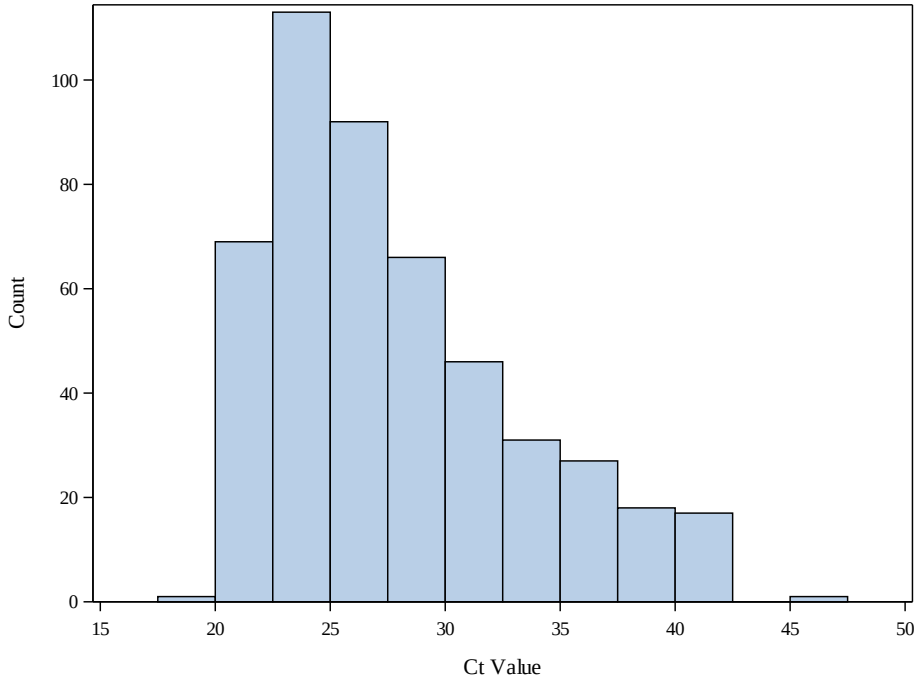


Figure 6 Cycle threshold distribution of NG positive specimens



Clinical reproducibility study results

A Reproducibility Study was performed across different sites, lots, operators/batches days, for **cobas**® CT/NG using three panels prepared from swabs and urine in **cobas**® PCR Media and cervical specimens in PreservCyt® Solution. PCR testing was performed at two external sites and one site that was in-house at Roche Molecular Systems. One panel consisted of the three sample matrices, with six concentrations per matrix, and three replicates per concentration for a total of 54 samples in one panel. A batch was comprised of one 54-sample panel and two controls (one positive control and one negative control). Two operators at each site tested one batch each per day. Two valid batches had to be completed within a 24-hour period. Each site received two of three reagent lots and performed 6 days of testing per reagent lot for a total of 12 days of testing.

The Reproducibility Study was executed with a total of 3,888 tests performed on the 6 panel groups, consisting of 1,296 tests for each panel type (urine, swab, and PreservCyt®), with only two failed tests each from PreservCyt®. No false positive results for either CT or NG were observed in the three panel types for negative panel members; thus the negative percent agreement was 100% for each analyte. Results for the positive panel members were highly reproducible across different lots, sites/instruments, days and operators/batches.

Negative panel results

For each sample type, all of the 216 valid tests from the negative panel members resulted in "Negative Results". Hence, for both CT and NG, the percent of correct results (analytical specificity) was estimated as 100% with a corresponding 95% exact confidence interval of 98.3%, 100% for **cobas**® PCR Media/urine, for **cobas**® PCR Media/swab and for PreservCyt®/cervical sample types.

Chlamydia trachomatis results

For each positive panel member, precision was evaluated using a random effects model by sample type with terms for lot, site, day, operator/batch within site, lot and day, and within-batch components on the corresponding analyte cycle threshold (Ct) values of **cobas**® CT/NG. Table 34 presents the total SD, and total percent CV (%) from these analyses for each panel type, respectively. The range of the total coefficient of variation, among positive panel members, was from 0.9% to 3.2%. The maximum total coefficient of variation was observed in the lowest concentration of positive panel members (0.3x LoD CT, 0.3x LoD NG) and most of that variability (98.6% for urine, 100% for swab and 81.7% for cervical) was explained by random error (within-batch).

Table 34 CT: overall mean, attributable percentage of total variance, total precision standard deviation, and CV(%) of **cobas**® CT/NG cycle threshold (Ct) values by CT positive panel member for each media type

Panel Member		Mean CT value		Percentage of Total Variance (CV[%])					Total Precision	
Media Type	Concen-tration	N[a]	Mean Estimate[b]	Site	Lot	Day	Operator /Batch	Within-Batch	SD[c]	CV(%)[d]
PCR Media/ Urine	0.3x LoD CT, 0.3x LoD NG	154	39.2	1.4% (0.4)	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	98.6% (3.0)	1.20	3.1
	1x LoD CT, Negative NG	216	36.8	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	100.0% (1.5)	0.54	1.5
	3x LoD CT, 1x LoD NG	216	35.4	2.4% (0.1)	0.0% (0.0)	21.1% (0.4)	0.0% (0.0)	76.5% (0.8)	0.33	0.9
	1x LoD CT, 3x LoD NG	216	36.9	0.0% (0.0)	0.0% (0.0)	10.3% (0.5)	4.4% (0.3)	85.3% (1.5)	0.59	1.6
PCR Media/ Swab	0.3x LoD CT, 0.3x LoD NG	128	39.5	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	100.0% (3.2)	1.26	3.2
	1x LoD CT, Negative NG	216	37.2	0.0% (0.0)	1.6% (0.2)	6.6% (0.5)	0.0% (0.0)	91.8% (1.7)	0.66	1.8
	3x LoD CT, 1x LOD NG	216	35.5	4.7% (0.2)	0.0% (0.0)	9.0% (0.3)	4.8% (0.2)	81.6% (0.9)	0.37	1.0
	1x LoD CT, 3x LoD NG	216	37.2	0.0% (0.0)	0.0% (0.0)	3.6% (0.4)	0.0% (0.0)	96.4% (2.3)	0.87	2.3
PreservCyt/ Cervical	0.3x LoD CT, 0.3x LoD NG	92	39.9	0.0% (0.0)	0.0% (0.0)	18.3% (1.4)	0.0% (0.0)	81.7% (2.9)	1.29	3.2
	1x LoD CT, Negative NG	216	37.0	12.0% (0.6)	1.9% (0.2)	0.0% (0.0)	0.0% (0.0)	86.2% (1.5)	0.60	1.6
	3x LoD CT, 1x LoD NG	216	35.6	0.6% (0.1)	3.7% (0.2)	0.0% (0.0)	6.3% (0.3)	89.3% (0.9)	0.36	1.0
	1x LoD CT, 3x LoD NG	214	36.8	13.1% (0.6)	3.7% (0.3)	5.3% (0.4)	2.3% (0.3)	75.6% (1.5)	0.63	1.7

Note: The table only includes results with detectable analyte. SD = standard deviation. CV(%) = percent coefficient of variation.

[a] Number of valid tests with detectable analyte.

[b] Calculated using SAS MIXED procedure.

[c] Calculated using the total variability from the SAS MIXED procedure.

[d] CV(%) = (SD/Mean) * 100.

LoD = Limit of Detection; CT = Chlamydia trachomatis; NG = Neisseria gonorrhoeae.

Table 35 through Table 37 present the percent agreement of CT test results for panel members by lot, site, and day for each media type, respectively.

Table 35 CT: Percent agreement by panel member for lot, site and day - **cobas®** PCR Media/urine

			CT Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site[b]**			**Day**		
Negative CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
0.3x LoD CT, 0.3x LoD NG	1.20	3.1	1	76.4	55/72	1	68.1	49/72	1	80.6	29/36
			2	70.8	51/72	2	73.6	53/72	2	77.8	28/36
			3	66.7	48/72	3	72.2	52/72	3	66.7	24/36
									4	77.8	28/36
									5	69.4	25/36
									6	55.6	20/36
1x LoD CT, Negative NG	0.54	1.5	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
Negative CT, 1x LoD NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
3x LoD CT, 1x LoD NG	0.33	0.9	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

			CT Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site[b]**			**Day**		
1x LoD CT, 3x LoD NG	0.59	1.6	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

[a] For CT Negative samples, Percent Agreement = (number of CT negative results/total valid results) x 100.
For CT Positive samples, Percent Agreement = (number of CT positive results/total valid results) x 100.
[b] Site 1, Site 2, and Site 3, respectively.
Ct = Cycle threshold; SD=Standard Deviation; CV = Coefficient of Variation; LoD = Limit of Detection.
CT = *Chlamydia trachomatis*; NG = *Neisseria gonorrhoeae*; n/a = not applicable.

Table 36 CT: Percent agreement by panel member for lot, site and day - **cobas®** PCR Media/swab

			CT Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site[b]**			**Day**		
Negative CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
0.3x LoD CT, 0.3x LoD NG	1.26	3.2	1	61.1	44/72	1	56.9	41/72	1	50.0	18/36
			2	59.7	43/72	2	61.1	44/72	2	63.9	23/36
			3	56.9	41/72	3	59.7	43/72	3	55.6	20/36
									4	61.1	22/36
									5	66.7	24/36
									6	58.3	21/36
1x LoD CT, Negative NG	0.66	1.8	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

			CT Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site[b]**			**Day**		
Negative CT, 1x LoD NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
3x LoD CT, 1x LoD NG	0.37	1.0	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
1x LoD CT, 3x LoD NG	0.87	2.3	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

[a] For CT Negative samples, Percent Agreement = (number of CT negative results/total valid results) x 100.
For CT Positive samples, Percent Agreement = (number of CT positive results/total valid results) x 100.
[b] Site 1, Site 2, and Site 3, respectively.
Ct = Cycle threshold; SD = Standard Deviation; CV = Coefficient of Variation; LoD = Limit of Detection.
CT = *Chlamydia trachomatis*; NG = *Neisseria gonorrhoeae*; n/a = not applicable.

Table 37 CT: Percent agreement by panel member for lot, site and day - PreservCyt®/cervical

			CT Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site[b]**			**Day**		
Negative CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
0.3x LoD CT, 0.3x LoD NG	1.29	3.2	1	38.9	28/72	1	34.7	25/72	1	40.0	14/35
			2	47.9	34/71	2	48.6	35/72	2	52.8	19/36
			3	41.7	30/72	3	45.1	32/71	3	38.9	14/36
									4	47.2	17/36
									5	41.7	15/36
									6	36.1	13/36
1x LoD CT, Negative NG	0.60	1.6	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
Negative CT, 1x LoD NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
3x LoD CT, 1x LoD NG	0.36	1.0	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

			CT Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site**[b]			**Day**		
1x LoD CT, 3x LoD NG	0.63	1.7	1	98.6	71/72	1	98.6	71/72	1	97.2	35/36
			2	100.0	71/71	2	100.0	71/71	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	35/35
									5	100.0	36/36
									6	100.0	36/36

[a] For CT Negative samples, Percent Agreement = (number of CT negative results/total valid results) x 100.
For CT Positive samples, Percent Agreement = (number of CT positive results/total valid results) x 100.
[b] Site 1, Site 2, and Site 3, respectively.
Ct = Cycle threshold; SD = Standard Deviation; CV = Coefficient of Variation; LoD = Limit of Detection.
CT = *Chlamydia trachomatis*; NG = *Neisseria gonorrhoeae*; n/a = not applicable.

Neisseria gonorrhoeae results

Analysis of variance components of the Ct values from valid NG test results were performed on positive panel members. Table 38 presents the total SD and total CV (%) from these analyses. The range of the total coefficient of variation, among positive panel members, was from 1.0% to 3.1%. The maximum total coefficient of variation was observed in the lowest concentration of positive panel members (0.3x LoD CT, 0.3x LoD NG) and most of that variability (98.7% for urine, 98.1% for swab and 85.3% for cervical) was explained by random error (within-batch).

Table 38 NG: overall mean, attributable percentage of total variance, total precision standard deviation, and CV(%) of **cobas**® CT/NG cycle threshold (Ct) values by NG positive panel member for each media type

Panel Member		**Mean CT value**		**Percentage of Total Variance (CV[%])**					**Total Precision**	
Media Type	**Concen-tration**	**N**[a]	**Mean Estimate**[b]	**Site**	**Lot**	**Day**	**Operator /Batch**	**Within-Batch**	**SD**[c]	**CV(%)**[d]
PCR Media/ Urine	0.3x LoD CT, 0.3x LoD NG	159	39.3	0.7% (0.3)	0.0% (0.0)	0.6% (0.2)	0.0% (0.0)	98.7% (3.0)	1.20	3.0
	Negative CT, 1x LoD NG	216	36.7	0.0% (0.0)	0.5% (0.1)	6.9% (0.5)	0.0% (0.0)	92.6% (1.7)	0.63	1.7
	3x LoD CT, 1x LoD NG	216	36.6	0.0% (0.0)	2.5% (0.3)	8.3% (0.5)	0.0% (0.0)	89.2% (1.6)	0.61	1.7
	1x LoD CT, 3x LoD NG	216	35.1	0.0% (0.0)	0.0% (0.0)	14.0% (0.4)	0.0% (0.0)	86.0% (1.0)	0.37	1.0
PCR Media/ Swab	0.3x LoD CT, 0.3x LoD NG	113	39.8	0.0% (0.0)	0.0% (0.0)	1.9% (0.4)	0.0% (0.0)	98.1% (3.1)	1.25	3.1
	Negative CT, 1x LoD NG	212	38.2	0.0% (0.0)	0.1% (0.1)	1.8% (0.4)	6.5% (0.7)	91.6% (2.6)	1.04	2.7
	3x LoD CT, 1x LoD NG	216	36.9	0.0% (0.0)	0.0% (0.0)	6.3% (0.6)	0.0% (0.0)	93.7% (2.1)	0.82	2.2

Panel Member		Mean CT value		Percentage of Total Variance (CV[%])					Total Precision	
Media Type	Concen-tration	N[a]	Mean Estimate[b]	Site	Lot	Day	Operator /Batch	Within-Batch	SD[c]	CV(%)[d]
PreservCyt® / Cervical	1x LoD CT, 3x LoD NG	216	35.7	0.0% (0.0)	3.8% (0.3)	14.4% (0.5)	0.0% (0.0)	81.8% (1.3)	0.50	1.4
	0.3x LoD CT, 0.3x LoD NG	112	39.5	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	14.7% (1.0)	85.3% (2.4)	1.04	2.6
	Negative CT, 1x LoD NG	216	35.7	7.2% (0.4)	4.9% (0.3)	0.0% (0.0)	0.0% (0.0)	87.9% (1.3)	0.49	1.4
	3x LoD CT, 1x LoD NG	216	36.3	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	9.6% (0.5)	90.4% (1.6)	0.61	1.7
	1x LoD CT, 3x LoD NG	215	34.6	2.3% (0.2)	0.0% (0.0)	5.8% (0.2)	12.0% (0.3)	79.8% (0.9)	0.34	1.0

Note: The table only includes results with detectable analyte. SD = standard deviation. CV(%) = percent coefficient of variation.

[a] Number of valid tests with detectable analyte.

[b] Calculated using SAS MIXED procedure.

[c] Calculated using the total variability from the SAS MIXED procedure.

[d] CV(%) = (SD/Mean) * 100.

LoD = Limit of Detection; CT = Chlamydia trachomatis; NG = Neisseria gonorrhoeae.

Panel Member			Percentage of Total Variance (CV[%])					Total Precision	
Media Type	Concentration	N[a]	Site	Lot	Day	Operator /Batch	Within-Batch	SD[b]	CV(%)[c]
PreservCyt/ Cervical	0.3x LoD CT, 0.3x LoD NG	112	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	14.7% (1.0)	85.3% (2.4)	1.04	2.6
	Negative CT, 1.0x LoD NG	216	7.2% (0.4)	4.9% (0.3)	0.0% (0.0)	0.0% (0.0)	87.9% (1.3)	0.49	1.4
	3.0x LoD CT, 1.0x LoD NG	216	0.0% (0.0)	0.0% (0.0)	0.0% (0.0)	9.6% (0.5)	90.4% (1.6)	0.61	1.7
	1.0x LoD CT, 3.0x LoD NG	215	2.3% (0.2)	0.0% (0.0)	5.8% (0.2)	12.0% (0.3)	79.8% (0.9)	0.34	1.0

Note: The table only includes results with detectable analyte. SD = standard deviation. CV(%) = percent coefficient of variation.

[a] Number of valid tests with detectable analyte.

[b] Calculated using the total variability from the SAS MIXED procedure.

[c] CV(%) = (SD/Mean)*100.

LoD = Limit of Detection; CT = *Chlamydia trachomatis*; NG = *Neisseria gonorrhoeae*.

Table 39 through Table 41 present the percent agreement of NG test results for panel members by lot, site, and day for each media type, respectively.

Table 39 NG: Percent agreement by panel member for lot, site and day - **cobas**® PCR Media/urine

			NG Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site**[b]			**Day**		
Negative CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
0.3x LoD CT, 0.3x LoD NG	1.20	3.0	1	79.2	57/72	1	70.8	51/72	1	77.8	28/36
			2	73.6	53/72	2	76.4	55/72	2	75.0	27/36
			3	68.1	49/72	3	73.6	53/72	3	72.2	26/36
									4	80.6	29/36
									5	61.1	22/36
									6	75.0	27/36
1x LoD CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
Negative CT, 1x LoD NG	0.63	1.7	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
3x LoD CT, 1x LoD NG	0.61	1.7	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

			NG Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site**[b]			**Day**		
1x LoD CT, 3x LoD NG	0.37	1.0	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

[a] For NG Negative samples, Percent Agreement = (number of NG negative results/total valid results) x 100.
For NG Positive samples, Percent Agreement = (number of NG positive results/total valid results) x 100.
[b] Site 1, Site 2, and Site 3, respectively.
Ct = Cycle threshold; SD = Standard Deviation; CV = Coefficient of Variation; LoD = Limit of Detection.
CT = *Chlamydia trachomatis*; NG = *Neisseria gonorrhoeae*; n/a = not applicable.

Table 40 NG: Percent agreement by panel member for lot, site and day - **cobas**® PCR Media/swab

			NG Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site**[b]			**Day**		
Negative CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
0.3x LoD CT, 0.3x LoD NG	1.25	3.1	1	50.0	36/72	1	50.0	36/72	1	52.8	19/36
			2	51.4	37/72	2	52.8	38/72	2	55.6	20/36
			3	55.6	40/72	3	54.2	39/72	3	44.4	16/36
									4	55.6	20/36
									5	52.8	19/36
									6	52.8	19/36
1x LoD CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
Negative CT, 1x LoD NG	1.04	2.7	1	100.0	72/72	1	97.2	70/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	94.4	68/72	3	97.2	70/72	3	97.2	35/36
									4	100.0	36/36
									5	97.2	35/36
									6	94.4	34/36
3x LoD CT, 1x LoD NG	0.82	2.2	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

			NG Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site[b]**			**Day**		
1x LoD CT, 3x LoD NG	0.50	1.4	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

[a] For NG Negative samples, Percent Agreement = (number of NG negative results/total valid results) x 100.
For NG Positive samples, Percent Agreement = (number of NG positive results/total valid results) x 100.
[b] Site 1, Site 2, and Site 3, respectively.
Ct = Cycle threshold; SD = Standard Deviation; CV = Coefficient of Variation; LoD = Limit of Detection.
CT = *Chlamydia trachomatis*; NG = *Neisseria gonorrhoeae*; n/a = not applicable.

Table 41 NG: Percent agreement by panel member for lot, site and day - PreservCyt®/cervical

			NG Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site[b]**			**Day**		
Negative CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
0.3x LoD CT, 0.3x LoD NG	1.04	2.6	1	63.9	46/72	1	59.7	43/72	1	54.3	19/35
			2	47.9	34/71	2	52.8	38/72	2	55.6	20/36
			3	44.4	32/72	3	43.7	31/71	3	47.2	17/36
									4	55.6	20/36
									5	52.8	19/36
									6	47.2	17/36
1x LoD CT, Negative NG	n/a	n/a	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36

			NG Percent Agreement[a]								
Panel Member	**Ct SD**	**Ct CV %**	**Lot**			**Site[b]**			**Day**		
Negative CT, 1x LoD NG	0.49	1.4	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
3x LoD CT, 1x LoD NG	0.61	1.7	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	72/72	2	100.0	72/72	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	36/36
									5	100.0	36/36
									6	100.0	36/36
1x LoD CT, 3x LoD NG	0.34	1.0	1	100.0	72/72	1	100.0	72/72	1	100.0	36/36
			2	100.0	71/71	2	100.0	71/71	2	100.0	36/36
			3	100.0	72/72	3	100.0	72/72	3	100.0	36/36
									4	100.0	35/35
									5	100.0	36/36
									6	100.0	36/36

[a] For NG Negative samples, Percent Agreement = (number of NG negative results/total valid results) x 100.
For NG Positive samples, Percent Agreement = (number of NG positive results/total valid results) x 100.
[b] Site 1, Site 2, and Site 3, respectively.
Ct = Cycle threshold; SD=Standard Deviation; CV = Coefficient of Variation; LoD = Limit of Detection.
CT = *Chlamydia trachomatis*; NG = *Neisseria gonorrhoeae*; n/a = not applicable.

Percentage agreement results

Table 42 shows the percent agreement with expected results for each target (CT, NG) with the associated 95% Exact CI.

Table 42 Percent agreement for panel members with concentration at or near the LoD (1x LoD) or 3x LoD

		CT		NG	
Media Type	**Panel Member**	**Percent Agreement**	**Percent Agreement 95% Exact CI**	**Percent Agreement**	**Percent Agreement 95% Exact CI**
PCR Media/Urine	1.0x LoD CT, Negative NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
	Negative CT, 1.0x LoD NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
	3.0x LoD CT, 1.0x LoD NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
	1.0x LoD CT, 3.0x LoD NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
PCR Media/Swab	1.0x LoD CT, Negative NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
	Negative CT, 1.0x LoD NG	100.0 (216/216)	(98.3, 100.0)	98.1 (212/216)	(95.3, 99.5)
	3.0x LoD CT, 1.0x LoD NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
	1.0x LoD CT, 3.0x LoD NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
PreservCyt®/ Cervical	1.0x LoD CT, Negative NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
	Negative CT, 1.0x LoD NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
	3.0x LoD CT, 1.0x LoD NG	100.0 (216/216)	(98.3, 100.0)	100.0 (216/216)	(98.3, 100.0)
	1.0x LoD CT, 3.0x LoD NG	99.5 (214/215)	(97.4, 100.0)	100.0 (215/215)	(98.3, 100.0)

Notes: LoD = Limit of Detection; CT = *Chlamydia trachomatis*; NG = *Neisseria gonorrhoeae*.
For panel members with concentrations at or near the limit of detection (e.g., 1x LoD) of the test, the lower limit of the 2-sided 95% exact CI of the percentage of correct test results should be equal to or greater than 91%.
For panel members with concentrations 3-times above the limit of detection (e.g., 3x LoD) of the test, the lower limit of the 2-sided 95% exact CI of the percentage of correct test results should be equal to or greater than 98%.

For panel members with concentrations at or near the limit of detection (e.g., 1x LoD) of the test, the lower limit of the 2-sided 95% exact CI of the percentage of correct test results was at least 97.4% for CT and 95.3% for NG.

For panel members with concentrations 3-times above the limit of detection (e.g., 3x LoD) of the test, the lower limit of the 2-sided 95% exact CI of the percentage of correct test results was 98.3% for both CT and NG.

Additional information

Key assay features

Sample types	• Endocervical swab collected in **cobas®** PCR Media • Vaginal swab collected in **cobas®** PCR Media • Self-collected Vaginal swab collected in **cobas®** PCR Media • Male and female urine stabilized in **cobas®** PCR Media • Cervical specimen collected in PreservCyt® Solution
Amount of sample required/processed	• ≥1000 µL required in sample tube for all swab samples, instrument processes 400 µL • ≥1000 µL required in sample tube for PreservCyt® samples, instrument processes 400 µL • ≥1200 µL required in sample tube for urine samples, instrument processes 850 µL
Test duration	• < 3.5 hours to first result

Symbols

The following symbols are used in labeling for Roche PCR diagnostic products.

Table 43 Symbols used in labeling for Roche PCR diagnostics products

	Ancillary Software		In Vitro diagnostic medical device
	Authorized representative in the European community		Lower Limit of Assigned Range
	Barcode Data Sheet		Manufacturer
	Batch code		Store in the dark
	Biological risks		Contains sufficient for <*n*> tests
	Catalogue number		Temperature limit
	Consult instructions for use		Test Definition File
	Contents of kit		Upper Limit of Assigned Range
	Distributed by		Use-by date
	For IVD performance evaluation only		Global Trade Item Number
	This product fulfills the requirements of the European Directive 98/79 EC for *in vitro* diagnostic medical devices.		

US Customer Technical Support 1-800-526-1247

Manufacturer and distributors

Table 44 Manufacturer and distributors

Roche Molecular Systems, Inc.
1080 US Highway 202 South
Branchburg, NJ 08876 USA
www.roche.com

Roche Diagnostics
9115 Hague Road
Indianapolis, IN 46250-0457 USA
(For Technical Assistance call the
Roche Response Center
toll-free: 1-800-526-1247)

Trademarks and patents

See http://www.roche-diagnostics.us/patents

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Document revision

Document Revision Information	
Doc Rev. 2.0 06/2019	Corrected manufacturer's address from Mannheim, Germany to Branchburg, NJ. Please contact your local Roche Representative if you have any questions.