Overview

Useful For

Detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* in non-FDA-approved specimen types

This test is **not intended for use** in medico-legal applications.

This test is **not useful for** the detection of *Chlamydia pneumoniae*.

Profile Information

Test ID	Reporting Name	Available Separately	Always Performed
MCRNA	C. trach, Misc, Amplified RNA	Yes	Yes
MGRNA	N. gonorr, Misc, Amplified RNA	Yes	Yes

Method Name

Transcription Mediated Amplification

NY State Available

Yes

Specimen

Specimen Type

Varies

Ordering Guidance

This test is used for specimens that **are not** FDA approved for this assay. Acceptable non-FDA-approved specimen types are ocular swabs, and peritoneal fluid. For FDA-approved specimen types, order CGRNA / *Chlamydia trachomatis* and *Neisseria gonorrhoeae*, Nucleic Acid Amplification, Varies.

Necessary Information

Specimen source is required.

Specimen Required

Swab specimens must be collected using an Aptima Collection Unisex Swab (T583), or Aptima Collection Multitest Swab (T584). These swabs are contained in the Aptima Collection Kit.

Submit only 1 of the following specimens:

Supplies:

Swab, Aptima Male/Female Collection (T583)

Swab, Aptima Multitest Swab Specimen Collection Kit (T584)

Specimen Type: Ocular (corneal/conjunctiva)

Container/Tube: Aptima Collection Multitest Swab or Aptima Swab Collection System

Specimen Volume: Swab

Collection Instructions:

1. Swab site using Aptima Collection Multitest Swab or Aptima Collection Unisex Swab.

Note: The white swab provided within the collection kit is a cleaning swab and should not be used for collection. Discard the white cleaning swab.

2. Place collection swab in transport tube provided in collection kit. Snap off swab at score line so swab fits into closed tube.

3. Cap tube securely and label tube with patient's entire name and collection date and time.

4. Transport swab container and store (refrigerated is preferred) within 60 days of collection.

Supplies: Aptima Thin Prep Transport Tube (T652)

Specimen Type: Peritoneal fluid (pelvic wash, cul-de-sac fluid)

Container/Tube: Aptima Specimen Transfer Tube

Specimen Volume: 1 mL

Collection Instructions:

1. Transfer specimen into the Aptima Specimen Transfer Tube within 24 hours of collection.

2. Cap tube securely and label tube with patient's entire name and collection date and time.

3. Transport Aptima Specimen Transfer Tube (refrigerated is preferred) within 30 days of collection.

Forms

If not ordering electronically, complete, print, and send a Microbiology Test Request (T244) with the specimen.

Specimen Minimum Volume

See Specimen Required

Reject Due To

All specimens will be evaluated at Mayo Clinic Laboratories for test suitability.

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Refrigerated (preferred)		APTIMA VIAL
	Ambient		APTIMA VIAL
	Frozen		APTIMA VIAL

Clinical and Interpretive

Clinical Information

Chlamydia is caused by the obligate intracellular bacterium *Chlamydia trachomatis* and is the most prevalent sexually transmitted bacterial infection in the United States.(1,2) In 2010, 1.3 million documented cases were reported to the CDC.(2) Given that 3 out of 4 infected women and 1 out of 2 infected men will be asymptomatic initially, the actual prevalence of disease is thought to be much greater than reported. The organism causes genitourinary infections in women and men and may be associated with dysuria as well as vaginal, urethral, or rectal discharge. In women, complications include pelvic inflammatory disease, salpingitis, and infertility. Approximately 25% to 30% of women who develop acute salpingitis become infertile.(2) Complications among men are rare but include epididymitis and sterility. Rarely, genital chlamydial infection can cause arthritis with associated skin lesions and ocular inflammation (Reiter syndrome). *C trachomatis* can be transmitted from the mother during delivery and is associated with conjunctivitis and pneumonia. Finally, *C trachomatis* may cause hepatitis and pharyngitis in adults.

Once detected, the infection is easily treated by a short course of antibiotic therapy. Annual chlamydia screening is now recommended for all sexually active women age 25 years and younger and for older women with risk factors for infection, such as a new sex partner or multiple sex partners. The CDC also recommends that all pregnant women be given a screening test for *Chlamydia* infection.(2) Repeat testing for test-of-cure is **not** recommended after treatment with a standard treatment regimen unless patient compliance is in question, reinfection is suspected, or the patient's symptoms persist. Repeat testing of pregnant women, 3 weeks after completion of therapy, is also recommended to ensure therapeutic cure.(2)

Gonorrhea is caused by the bacterium *Neisseria gonorrhoeae*. It is also a very common sexually transmitted infection (STI), with 301,174 cases of gonorrhea reported to CDC in 2009.(2,3) Many infections in women are asymptomatic and the true prevalence of gonorrhea is likely much higher than reported. The organism causes genitourinary infections in women and men and may be associated with dysuria as well as vaginal, urethral, or rectal discharge. Complications include pelvic inflammatory disease in women and gonococcal epididymitis and prostatitis in men. Gonococcal bacteremia, pharyngitis, and arthritis may also occur. Infection in men is typically associated with symptoms that would prompt clinical evaluation. Given the risk for asymptomatic infection in women, screening is recommended for women at increased risk of infection (eg, women with previous gonorrhea or other STI, inconsistent condom use, new or multiple sex partners, and women in certain demographic groups such as those in communities with high STI prevalence).(2,3) The CDC currently recommends dual antibiotic treatment due to emerging antimicrobial resistance.(2)

Culture was previously considered to be the gold standard test for diagnosis of *C trachomatis* and *N gonorrhoeae* infections.(2) However, organisms are labile in vitro, therefore, precise specimen collection, transportation, and processing conditions are required to maintain organism viability, which is necessary for successful culturing. In comparison, nucleic acid amplification testing (NAAT) provides superior sensitivity and specificity and is now the recommended method for diagnosis in most cases.(4-6) Immunoassays and non-amplification DNA tests are also available for *C trachomatis* and *N gonorrhoeae* detection, but these methods are significantly less sensitive and less specific than NAAT.(2)

Improved screening rates and increased sensitivity of NAAT testing have resulted in an increased number of

accurately diagnosed cases of both chlamydia and gonorrhea.(2-6) Improved detection rates result from both the increased performance of the assay and the patients' easy acceptance of urine testing. Early identification of infection enables sexual partners to seek testing and/or treatment as soon as possible and reduces the risk of disease spread. Prompt treatment reduces the risk of infertility in women.

Reference Values

Negative

Interpretation

A positive result indicates that rRNA of *Chlamydia trachomatis* and/or *Neisseria gonorrhoeae* is present in the specimen tested and strongly supports a diagnosis of chlamydial/gonorrheal infection.

A negative result indicates that rRNA for *C trachomatis* and/or *N gonorrhoeae* was not detected in the specimen.

The predictive value of an assay depends on the prevalence of the disease in any particular population. In settings with a high prevalence of sexually transmitted disease, positive assay results have a high likelihood of being true positives. In settings with a low prevalence of sexually transmitted disease, or in any setting in which a patient's clinical signs and symptoms or risk factors are inconsistent with gonococcal or chlamydial urogenital infection, positive results should be carefully assessed and the patient retested by other methods (eg, culture for *N gonorrhoeae*), if appropriate.

A negative result does not exclude the possibility of infection. If clinical indications strongly suggest gonococcal or chlamydial infection, additional specimens should be collected for testing. A result of indeterminate indicates that a new specimen should be collected.

This test has not been shown to cross react with commensal (nonpathogenic) *Neisseria* species present in the oropharynx.

Cautions

This report is intended for use in clinical monitoring or management of patients; it is not intended for use in medico-legal applications.

Appropriate specimen collection and handling is necessary for optimal assay performance.

Results should be interpreted in conjunction with other laboratory and clinical information.

A negative test result does not exclude the possibility of infection. Improper specimen collection, concurrent antibiotic therapy, presence of inhibitors, or low numbers of organisms in the specimen (ie, below the sensitivity of the test) may cause false-negative test results.

In low-prevalence populations, positive results must be interpreted carefully as false-positive results may occur more frequently than true-positive results in this setting.

In general, this assay should not be used to assess therapeutic success or failure, since nucleic acids from these organisms may persist for 3 weeks or more following antimicrobial therapy.

No interference is expected with swab specimens due to:

-Blood

-Lubricants and spermicides

This assay **does not** detect *Chlamydia pneumoniae*.

Supportive Data

Chlamydia trachomatis

Accuracy:

1. Clinical Specimens

Non-FDA approved specimen types were collected in Hologic (GEN-PROBE) APTIMA collection devices according to the manufacturer's instructions and tested using the APTIMA Combo 2 assay on the Tigris DTS System. Results were compared to those obtained by other CLIA-certified laboratories using the Tigris system. All specimens were within product insert stability requirements at the time of testing on the Tigris system. Clinical specimens were stored frozen until the time of testing.

Non-FDA Approved Sources for detection of *C trachomatis* (see additional spiking data below):

Peritoneal fluid		**Reference result**		
Aptima Unisex*		Positive	Negative	Total
APTIMA (Mayo)	Positive	0	0	0
	Negative	0	10	10
	Total	0	10	10

*Additional studies using raw peritoneal fluid in lieu of swab collection were performed for a more concise specimen collection and enhanced recovery. One mL of peritoneal fluid was added to a Hologic APTIMA specimen transfer kit then spiked with *C trachomatis* and *Neisseria gonorrhoeae* at the limit of detection.

Source	**Positives**	**CT IFU/assay**	**NG cfu/assay**	**Concordance**
Peritoneal fluid	20	3	50	100%

IFU-inclusion forming units

2. Spiked Specimens:

Analyte-negative ocular (n=32)and peritoneal fluid (n=30) specimens were spiked at the approximate limit of detection (LOD) and tested to supplement clinical specimen validation data (see analytical sensitivity validation data below).

Percent concordance by specimen type:

Source	**Positive (number tested)**	**Negative (number tested)**	**Concordance**
Peritoneal Fluid	30 (30)	0 (0)	100%
Ocular	32 (32)	35 (35)	100%

3. Total Accuracy (Clinical and spiked specimens combined):

Source	Collection device*	Total tested**		Sensitivity	Specificity
		Positive (number tested)	Negative (number tested)		
Peritoneal fluid	**Unisex**	30 (30)	10 (10)	100%	100%
Ocular	**Unisex**	32 (32)	35 (35)	100%	100%

*****All collection devices are manufactured by Hologic (GEN-PROBE) for use with the Aptima assay.

**Positive and negative status are based on the result by the comparator method (clinical specimens) or expected result (spiked specimens).

Analytical Sensitivity/LOD:

The LOD of this assay was established at 3 IFU (inclusion forming units) using a quantified whole organism control from Zeptometrix. The LOD was confirmed in all non-FDA approved specimens that will be accepted for testing with this assay (, ocular specimens). At least 30 clinical specimens of each source grouping were spiked with *C trachomatis* at 3 IFU/assay. Results are as follows:

Specimen type	LOD (IFU)	Number positive (number tested)	% Positive
Ocular	3	32 (32)	100
Peritoneal fluid	3	30 (30)	100

According to the package insert, the LOD for detection of *C trachomatis* using FDA approved specimens and collection devices is 1 IFU per assay, with a range of 3 to 0.1 IFU per assay. Therefore, the LOD for non-FDA approved specimens falls within this range and is considered acceptable.

Analytical Specificity:

To augment the specificity panel performed by Hologic (GEN-PROBE) as outlined in the APTIMA product insert, an additional panel was tested by the Tigris DTS System using the APTIMA COMBO 2 Assay. Negative patient matrix specimens collected in Hologic collection devices were spiked with specificity panel organisms and tested. Organisms were chosen based on their ability to cause disease similar to *C trachomatis*, or be normal flora in non-FDA approved specimen sources. The assay did not cross-react with any members of the specificity panel.

N gonorrhoeae

Accuracy:

1. Clinical Specimens:

Non-FDA approved specimen types were collected in Hologic (GEN-PROBE) APTIMA collection devices according to the manufacturer's instructions and tested using the APTIMA Combo 2 assay on the Tigris DTS System. Results were compared to those obtained by other CLIA-certified laboratories using the Tigris system. All specimens were within product insert stability requirements at the time of testing on the Tigris system. Clinical specimens were stored frozen until the time of testing.

Non-FDA Approved Sources for detection of *N gonorrhoeae* (see additional spiking studies below):

Peritoneal Fluid		Reference Result		
Aptima Vaginal Self Collect Kit*		Positive	Negative	Total
APTIMA (Mayo)	Positive	0	0	0
	Negative	0	10	10
	Total	0	10	10

*Additional studies using raw peritoneal fluid in lieu of swab collection were performed for a more concise specimen collection and enhanced recovery. One mL of peritoneal fluid was added to a APTIMA specimen transfer kit then spiked with *C trachomatis* and *N gonorrhoeae* at the limit of detection.

Source	Positive	CT IFU/assay	NG CFU/assay	Concordance
Peritoneal fluid	20	3	50	100%

2. Spiked Specimens:

Negative specimens were spiked at the approximate limit of detection (LoD) and tested to supplement clinical specimen validation data (see analytical sensitivity validation data below).

Source	Positive (number tested)	Negative (number tested)	Concordance
Peritoneal Fluid	30 (30)	0 (0)	100%
Ocular	30 (30)	46 (46)	100%

3. Total Accuracy (Clinical and spiked specimens combined):

Source	Collection device*	Total tested**		Sensitivity	Specificity
		Positive (number tested)	Negative (number tested)		
Peritoneal fluid	Vaginal Self Collect	30 (30)	10 (10)	100%	100%
Ocular	Unisex	30 (30)	46 (46)	100%	100%

*All collection devices are manufactured by Hologic for use with the Aptima assay

**Positive and negative status are based on the result by the comparator method (clinical specimens) or expected result (spiked specimens)

Analytical Sensitivity (LOD)

The LOD was established by preparing dilutions of *N gonorrhoeae* (ATCC strain 43069). The LOD was determined to be 12.5 CFU (colony forming units)/assay. Although specimens diluted to a final concentration of 12.5 CFU/assay gave 100% positive results, we chose only to test the analytical sensitivity claim in the product insert, which is 50 CFU/assay. The LOD was confirmed in all non-FDA approved specimens that will be accepted for testing with this assay (ocular specimens). Clinical specimens of each source/grouping were spiked with *N gonorrhoeae* at 50 CFU/assay and tested with positive and negative controls as per standard protocol.

Summary of Results:

Specimen type	Limit of detection	Number positive (number tested)	% Positive
Ocular	50 CFU/assay	30 (30)	100
Peritoneal fluid	50 CFU/assay	30 (30)	100

Analytical Specificity:

To augment the specificity panel performed by Hologic as outlined in the APTIMA product insert, an additional panel was tested by the Tigris DTS System using the APTIMA COMBO 2 Assay. Analyte-negative patient specimens collected in Hologic collection devices were spiked with specificity panel organisms and tested. Organisms were chosen based on their ability to cause disease similar to *N gonorrhoeae* or be normal flora in non-FDA approved specimen sources. The assay did not cross-react with any members of the specificity panel.

Clinical Reference

1. Centers for Disease Control and Prevention. 2014. Recommendations for the laboratory-based detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae*, 2014. MMWR Morb Mortal Wkly Rep. 2014;63:1-18

2. Centers for Disease Control and Prevention: Sexually Transmitted Diseases Treatment Guidelines, 2015. MMWR Morb Mortal Wkly Rep. 2015 Jun 5;64(RR-03):1-137.

3. Centers for Disease Control and Prevention. 2002. Reporting of laboratory-confirmed chlamydial infection and gonorrhea by providers affiliated with three large Managed Care Organizations-United States, 1995-1999. MMWR Morb Mortal Wkly Rep. 2002;51:256-259

4. Crotchfelt KA, Pare B, Gaydos C, Quinn TC: Detection of *Chlamydia trachomatis* by the GEN-PROBE AMPLIFIED *Chlamydia trachomatis* Assay (AMP CT) in urine specimens from men and women and endocervical specimens from women. J Clin Microbiol. 1998 Feb;36(2):391-394

5. Gaydos CA, Quinn TC, Willis D, et al: Performance of the APTIMA Combo 2 assay for detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* in female urine and endocervical swab specimens. J Clin Microbiol. 2003 Jan;41(1):304-309

6. Chernesky MA, Jang DE: APTIMA transcription-mediated amplification assays for *Chlamydia trachomatis* and *Neisseria gonorrhoeae*. Expert Rev Mol Diagn. 2006 Jul;6(4):519-525

Performance

Method Description

The Hologic Aptima Combo 2 Assay combines the technologies of target capture, transcription-mediated amplification, and dual kinetic assay. The detection of the rRNA amplification product sequences (amplicon) is achieved using nucleic acid hybridization. Single-stranded chemiluminescent DNA probes are labeled and combine with amplicon to form stable RNA:DNA hybrids. Light emitted from the labeled RNA:DNA hybrids is measured as photon signals in a luminometer.(Package insert: Hologic APTIMA Combo 2 Assay. 502446 Hologic, Inc; Rev. 005 06/2018)

PDF Report

No

Day(s) Performed

Monday through Saturday

Report Available

1 to 4 days

Performing Laboratory Location

Rochester

Fees and Codes

Fees

- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their Regional Manager. For assistance, contact Customer Service.

Test Classification

This test has been modified from the manufacturer's instructions. Its performance characteristics were determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information

MCRNA-87491

MGRNA-87591

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
MCTGC	Misc C trach/N gonor Amplified RNA	In Process

Result ID	Test Result Name	Result LOINC Value
SRC11	SOURCE:	31208-2
SRC22	SOURCE:	31208-2
34507	C. trach, Misc, Amplified RNA	43304-5
34508	N. gonorr, Misc, Amplified RNA	43305-2