

# CHLAMYDIA TRACHOMATIS

For in *Vitro* diagnostic use only

## Rapid immunochromatographic test for qualitative detection of Chlamydia trachomatis

### I. INTRODUCTION AND INTENDED USE

The Chlamydia Trachomatis Test is intended for in vitro diagnostic use in the rapid, qualitative manual detection of Chlamydia trachomatis directly from female endocervical cells, male urethral cells and male urine specimens from symptomatic patients. The test is intended for use as an aid in the diagnosis of Chlamydia infections.

Chlamydia includes three species: Chlamydia trachomatis, Chlamydia pneumonia and Chlamydia psittaci. Chlamydia trachomatis, a human pathogen, is subdivided into 15 known serovars; 3 are associated with Lymphogranuloma Venereum and the remaining 12 with trachomatis and genitourinary infection. Common complications of women infected with Chlamydia include cervicitis, urethritis, endometritis, pelvic inflammatory disease and increased incidence of ectopic pregnancy and infertility, and of men include urethritis and epididymitis. Chlamydia trachomatis infection has a high prevalence and asymptomatic carriage rate. Approximately 70% of women with endocervical infections and up to 50% of men with urethral infections are asymptomatic.

Chlamydia psittaci, mainly an animal pathogen, is associated with respiratory disease caused by exposure to infected birds and not transmitted through human interaction.

Chlamydia pneumonia, first isolated in 1983, is a human pathogen and is associated with respiratory infections and pneumonia.

Traditionally, Chlamydia infection is diagnosed by detection of Chlamydia inclusions in tissue culture cells.

Although this method is the most sensitive and specific laboratory method, it is costly, labor intensive, time consuming (48-72 hours) and not routinely available in most institutions. Direct immunofluorescence assay requires specialized equipment and a skilled operator to interpret results. The Chlamydia Trachomatis Test, utilizing the gold particle based immunoassay, provides a simple, rapid, specific yet highly sensitive method of detection of Chlamydia antigen.

### II. PRINCIPLE

The Chlamydia Trachomatis Test is a rapid qualitative immunoassay based on the immunochromatographic principle. (In the assay procedure, a clinical specimen is obtained and placed into an extraction tube containing Extraction Solution A. After two minutes. Extraction Solution B is added to the tube. 3 drops (approximately 150µl) of extracted sample is added to the sample well). The membrane is pre-coated with anti-genus specific lipopolysaccharide (LPS) monoclonal antibody on the test band (T) region and goat anti-mouse antibody on the control band (C) region. During testing, the sample is allowed to react with the colloidal gold particles which have been coated with monoclonal anti-chlamydia antibody, and then migrates laterally across the membrane by capillary action. If the sample contains Chlamydia antigen, a colored band with a specific antibody- Chlamydia antibody-colloidal gold particle complex will form on the membrane in the test band (T) region. If Chlamydia antigen is not present, a pink line will only form on control band (C) region. To serve as a procedural control, a colored band at the control band (C) region will always appear regardless of the presence of Chlamydia antigen.

### III. COMPOSITION OF CHLAMYDIA TRACHOMATIS KIT

Each kit contains:

1. Chlamydia trachomatis Test Card	20 items
2. EXTR. SOL. A in plastic dropper bottles containing 0.2 M NaOH	1 x 7,5 mL
3. EXTR. SOL. B in plastic dropper bottles containing 0.2 M HCl	1 x 7,5 mL
4. Extraction tube with filter caps	20 items
5. Instructions leaflet	1 item

### **AUXILIARY MATERIALS (Not supplied with this kit)**

- Specimen collection container
- Sterile swabs (swabs with rayon or dracon tips)
- Timer

### IV. STORAGE AND STABILITY

1. Kit components should be stored at room temperature (between +4°C and +30°C).
2. Do not freeze the test kit.
3. Chlamydia Trachomatis card is stable until the expiry date stated on the package label.

### V. PRECAUTIONS

- For professional and *IN VITRO* diagnostic use only.
- Do not use any of the kit contents after the expiration date. Do not mix kit components from different lots. Do not mix reagent bottle caps.
- Use appropriate precautions in the collection, handling, storage and disposal of specimens, device and used kit contents. All specimens, reagents and controls should be handled as if they contain infectious agents. When the assay procedure is completed, dispose of used swabs carefully after autoclaving them at 121°C for over 20 minutes or pre-treating them with 0.5%-1% sodium hypochlorite (or household bleach) for an hour.
- Extraction Solution A contains sodium hydroxide (a basic solution) and Extraction Solution B contains hydrochloric acid (an acid solution). If either of the solutions contacts the skin or eye, flush with plenty of water.
- Use sterile swabs or cytology brushes to obtain endocervical specimens.
- Do not eat, drink or smoke in the area where specimens and kit reagent are handled. Wear protective clothing such as laboratory coats and disposable gloves while collecting and assaying samples and device.
- Extraction Solution contains sodium azide which may react with lead or copper plumbing to form potentially explosive metal azides. When disposing of buffered saline or extracted samples, always flush with copious quantities of water to prevent azide build up
- Not follow the operation procedure/violation, abbreviation of instruction procedure, failure of the device or failure to correctly interpret of the results may lead to incorrect patient management decision and inappropriate health response. In the context of individual patient management, a false negative report could lead to delays in providing (or failure to provide) definitive diagnosis and appropriate treatment and infection control and prevention measures. If untreated, chlamydia infection may persist with the consequence of development of PID that can lead to infertility. In the pregnant women the untreated infection can cause a conjunctivitis and pneumonia to infant. The conjunctivitis can lead to blind in the infant. A false positive report could lead to unnecessary or inappropriate treatment or unnecessary control and prevention action. In the worst case, the consequence of untreated Chlamydia infection can cause permanent damage.



### VI. SPECIMEN COLLECTION AND PREPARATION

For the best performance of any Chlamydia trachomatis test, an accurate sample collection technique is extremely important.

#### A) Female endocervical specimens:

1. Before specimen collection, use a separate swab or cotton ball to remove excess mucus from the endocervical area and discard.

**Note: failure to remove mucus may result in false-positive results.**

2. *Insert the swab* into the endocervical canal past the squamocolumnar junction, until most of the tip is no longer visible. This will permit acquisition of columnar or cuboidal epithelial cells which are the main reservoir of Chlamydia organism. Rotate the swab for 15-20 seconds, withdraw it without touching any vaginal surface.
3. Alternatively, endocervical specimens can be collected with a cytology brush (Not provided. Caution; do not use cytology brushes with pregnant patients). Insert the cytology brush into the endocervical canal past the squamocolumnar junction. Leave in place two to three seconds. Rotate the cytology brush two full turns, and then withdraw the brush without touching any vaginal surface.
4. Place the swab in the extraction tube, if the test is to be conducted immediately.

#### B) Male Urethral specimens :

1. Use standard wire-shafted fiber-tipped swabs (not provided) for urethral specimen collection. Instruct the patients not to urinate at least one hour prior to specimen collection.
2. Insert the swab into the urethra about 2-4 cm, rotate for 3-5 seconds and withdraw it.
3. Place the swab to the extraction tube, if the test is to be conducted immediately.
4. Do not place the swab in any transport device containing medium since transport medium interferes with the assay.

If immediate testing is not possible, the patient sample should be placed in a dry transport tube for storage or transport. The swabs may be stored for 4 hours at room temperature (10-30°C) or 24 hours at refrigerated at (4-8°C). Do not freeze. All specimens should be allowed to reach a room temperature of 10-30°C before testing.

#### C) Male Urine specimens :

1. Collect 15-30 mL of first urine in the morning in a clean dry container.
2. Shake and mix the urine sample evenly, extract 10 mL into a centrifuge tube, add 10 mL distilled water, and centrifuge for 15-20 minutes at 3000 rpm. Carefully discard the supernatant. Turn the centrifuge tube upside down and suck the liquid on the edge of the centrifuge tube with absorbent paper.
3. If necessary, centrifuge the precipitate with distilled water again according to the above conditions, and the precipitate will be used for detection.
4. If immediate testing is not possible, the urine sample can be refrigerated for less than 24 hours at 2-8°C.

### VII. ASSAY PROCEDURE

Review "specimen collection" instructions. Do not open pouches until ready to perform the assay. Make sure that all reagents, test units and swabs, are at room temperature before beginning the assay. To avoid cross contamination, do not allow the tip of the reagent bottle to come in contact with sample swabs or Extraction tubes.

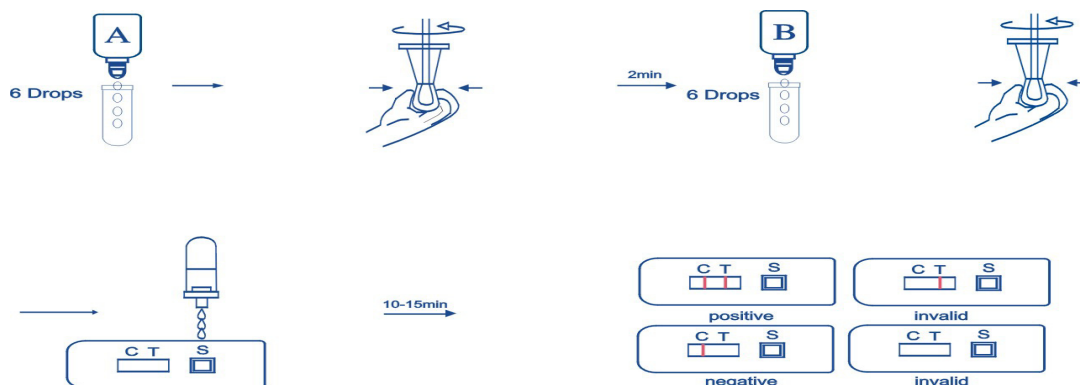
#### A) SPECIMENS EXTRACTION:

##### Preparation of Endocervical and/or Urethral swab specimens or urine sediment

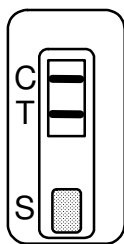
- **Swab:** Place a new Extraction tube in the designated area of the workstation. Add 6 drops of Extraction Solution A to Extraction tube.
  - Immerse the patient's swab into the Extraction tube, and extract 2 minutes at room temperature. During extraction, use a circular motion to roll the swab against the side of the Extraction tube so that the liquid is expressed from the swab and reabsorbed.
  - At the end of the extraction time, add 6 drops of solution B. Squeeze the swab firmly against the tube to expel as much liquid as possible from the swab. Discard the swab following guidelines for handling infectious agents.
  - **Sediment:** add 6 drops of Extraction Solution A directly to the centrifuge tube and shake well.
  - Pour the solution to the extraction tube, than add 6 drops of Extraction Solution B.
- The extracted specimen can remain at room temperature for 60 minutes without affecting the result of the Chlamydia Test.

#### B) TEST PROCEDURE

- Follow instructions for specimen collection and extraction
- Remove Chlamydia Trachomatis card from its protective pouch and place on a level surface, clean and dry. Label the device with patient's name or control number.
- Place the cap on the extraction tube. Add 3 drops (approx. 150 µl) of extracted sample from Extraction tube to the sample well.
- Wait for test band (S) to appear. The test results should be read in 10 minutes after adding the extracted specimen to the sample well. Depending on the amount of Chlamydia antigen organisms on the swab, positive result may be visible as soon as 1 minute. However, to confirm a negative result the complete reaction time of 15 minutes is required. **Do not interpret result after 15 minutes.**

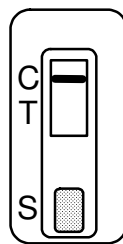


### VIII READING TEST RESULTS



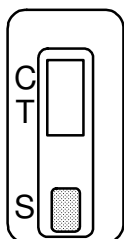
#### Positive

Two colored lines should be observed in the viewing window. The line in the test region (T) is the probe line; the line in the control region (C) is the control line, which is used to indicate proper performance of the device. **The color intensity of the test line may be weaker or stronger than that of the control line.**



#### Negative

The control line (C) appears in the test window, but the test line (T) is not visible.



#### Invalid

No line appears in the control region. Under no circumstances should a positive sample be identified until the control line (C) forms in the viewing area. If the control line does not form, the test result is inconclusive and the assay should be repeated.

### IX. DISPOSAL

- Return test device and droppers to original pouches.
- Dispose of test materials according to local, state and federal regulations.

### X. PERFORMANCES CHARACTERISTICS

#### A) Accuracy

A study was performed on 172 clinical samples (from 3 different clinics, including male and female swab samples and male urine) assayed using Chlamydia Trachomatis card and Clearview Chlamydia MF test (extended Golden Standard). The results were read at 10-15 minutes.

		Clearview Chlamydia MF		Total
		+	-	
Chlamydia trachomatis test	+	74	3	77
	-	2	93	95
Total		76	96	172

Relative Sensitivity: 97.37%

Relative Specificity: 96.88%

Relative Accuracy: 97.1%

#### B) Specificity

The antibody used in Chlamydia Trachomatis Test has been shown to detect all 15 Chlamydia serovars. In addition, Chlamydia psittaci and Chlamydia pneumonia strains have been tested with the Chlamydia Trachomatis Test and gave a positive result.

Cross reactivity with other organisms has been studied using suspensions of 10<sup>6</sup>CFU/ml. The following organisms were not detected using this Chlamydia Test:

Acinetobacter	Candida albicans
Salmonella typhi	Escherichia coli
Staphylococcus aureus	Streptococcus faecalis
Neisseria gonorrhoeae	Streptococcus faecium
Pseudomonas	Trichomonas vaginalis
Hemoglobin	Spermicides
Gynecological Lubrificants	Talcum powder

#### C) Sensitivity

A study was performed to verify the sensitivity of Chlamydia Trachomatis card. A commercially available Chlamydia Test (Clearview Chlamydia MF) was tested side by side using different levels of Chlamydia controls of some serovars (from 0 at 15000 IFU/mL). The results indicated that the sensitivity of the Chlamydia Trachomatis card is comparable to the commercially available product and is 4000 IFU/mL.

### XI. QUALITY CONTROL

Chlamydia Trachomatis card includes a procedure control. A pink colored band appearing in the control band (C) region of the membrane indicates proper performance and reactive reagents.

### XII. LIMITATIONS













1. The Chlamydia Trachomatis card test is for in vitro diagnostic use only. The test should be used for the detection of Chlamydia trachomatis antigen only and for specimens collected from the endocervical or urethral regions or from male urine.
2. Specimen with an excessive amount of mucus or blood may give false positive results.



3. Like all immunological tests for *Chlamydia trachomatis*, this test cannot distinguish between biologically active and inactive organisms.
4. Chlamydia Trachomatis card does not specifically differentiate *C. trachomatis*, *C. Pneumonia* or *C. Psittaci*. Detection of Chlamydia is depended on the number of organisms present in the specimen. This may be affected by specimen collection methods and patient factors such as age, history of Sexually Transmitted Disease, presence of symptoms, etc.
5. This test will only indicate the presence or absence of Chlamydia trachomatis antigen in the specimens and should not be used as the only basis for the diagnosis of chlamydial infection.
6. If clinical symptoms persist and the test result is negative, additional follow-up testing with e.g. cell culture method is required.
7. As for all diagnostic procedures, a diagnosis should not be based on a single test result but, should only be made by a physician after pooling the laboratory results, clinical investigations and patient's history.

### XIII. BIBLIOGRAPHY

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4. Global incidence and prevalence of selected curable sexually transmitted infections-2008 (W.H.O.)
5. Draft guidance for industry-Devices for Chlamydia (FDA may 2011)
6. Chlamydia (Istituto superiore di sanità-Italian MOH)-italian language
7. Infection by Chlamydia trachomatis (Società interdisciplinare per lo studio delle malattie sessualmente trasmesse)
8. Chlamydiacea (Università degli studi di Perugia-Dipartimento di Medicina e chirurgia Anni 2011-2012)


 IVD	In Vitro Diagnostic Medical Device		Temperature limitation	 LOT	Batch code (EXXX)		Manufacturer		Keep dry		Non-sterile
	Consult Instructions for use		Use by (year/month)	 REF	Catalogue number		Do not reuse		Fragile, handle with care		Keep away from heat

### CONTENTS

1. Chlamydia trachomatis Test Card
2. EXTR. SOL. A in plastic dropper bottles containing 0.2 M sodium hydroxide
3. EXTR. SOL. B in plastic dropper bottles containing 0.2 M hydrochloride acid
4. Extraction tube with filter caps
5. Instructions for use

### REF VQ81406 (20 test)

- 20 items
- 1 x 7,5 mL
- 1 x 7,5 mL
- 20 items
- 1 item

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