For urethral specimens:

1. Immerse the patient’s swab or the brush in the extraction tube and extract the chlamydia antigen according to the specimen type.
2. At the end of the extraction time, add 5 drops of the extraction solution B to the test cassette without additional incubation time.
3. After opening the foil pouch, place the test device into the plastic holder. Add 5 ml of extraction solution B to the test tube. Transfer the suspension into the extraction tube with a disposable pipette, and mix the solution with the swab or brush. Then press the swab/brush firmly against the tube in order to squeeze as much liquid as possible into the swab or brush. Discard the swab/brush in accordance with guidelines for handling infectious materials.
4. The specimens extracted can be kept at room temperature for 60 minutes without affecting the results of the chlamydia test. Prepare male urine:
   - The urine specimens should be centrifuged in order to collect all the particles which may contain chlamydia cells. Centrifuge the urine (at least 15 min) at 10,000 rpm for 10 minutes.
   - Carefully drain the excess and add 5 drops of extraction solution A to the tube, re-suspend the pellet with a disposable pipette and incubate at room temperature for 2 minutes.
   - Transfer the suspension into the extraction tube with a disposable pipette, add 5 drops of extraction solution B and mix (e.g. by means of bringing the solution up into a pipette repeatedly). The sample can now be placed in the test cassette without additional incubation time.

For professional in vitro diagnostic use only

**PRINCIPLE**

The **Linear Chlamydia Test Device (Swab/Urine)** is a rapid immunochromatographic test. In the test procedure a clinical specimen is obtained and placed in an extraction tube with extraction solution A. After 2 minutes extraction solution B is added to the tube. After extraction 3 drops (approximately 120 µl) of the extracted sample are added to the test cassette sample well. In the test the membrane was coated with antigen-specific monoclonal antibodies on the test line and with a goat anti-rabbit antibody on the control line. During testing any antigens present react with the gold-marked monoclonal antibody and then move laterally on the membrane by capillary action. If the sample contains chlamydia antigens, a coloured line with a specific antibody-chlamydia-antibody gold particle complex will form on the membrane in the test band region. If no chlamydia antigen is present, only a pink line appears in the control zone, whether chlamydia is present or not.

**REAGENT COMPOSITION**

- Chlamydia antibody coated on the membrane.
- Antibody-chlamydia-antibody gold particle complex.
- The chlamydia antigen test is intended for in vitro diagnostic use only.

**PACKAGING CONTENTS**

- 20 tests
- 20 test cassettes
- 20 Sterilized swabs with Dacron tips (CED482)
- 20 Extraction tube and dropper cap
- Reagent A (0.2M NaOH). Xi: irritant; R 36/38 irritates eyes and skin.
- Workstation
- Reagent B (0.2N HCl)
- Quantitative pipette

**MATERIALS REQUIRED**

- Cytology brushes for the optimum taking of samples from cervical swabs
- Special swabs for taking samples from the urethra
- Stopwatch
- For urine samples: Urine-storage vessel, one-way pipette and centrifuge
- For taking samples from the urethra: Special swabs
- Positive and negative controls

**PROCEDURE**

Allow the test, specimen, reagents, and/or controls to reach room temperature (15-30°C) prior to testing.

1. Remove the test device from the sealed foil pouch and use it as soon as possible. Best results will be obtained if the test is performed immediately after opening the foil pouch.
2. Extract the Chlamydia antigen according to the specimen type.

**Specimens and control extraction:**

Prepare endocervical or urethral specimens:

- Place a clean, identified extraction tube into the plastic holder. Add 5 drops of extraction solution A to the extraction tube.
- Immerse the patient’s swab or the brush in the extraction tube and extract for 2 minutes at room temperature. During extraction, use a circular motion to roll the swab/brush against the side of the extraction tube so that the liquid is expressed from the swab/brush and can be reabsorbed.
- At the end of the extraction time, add 5 drops of the extraction solution B and mix the solution with the swab or brush. Then press the swab or brush firmly against the tube in order to squeeze as much liquid as possible out of the swab or brush. Discard the swab/brush in accordance with guidelines for handling infectious materials.
- The specimens extracted can be kept at room temperature for 60 minutes without affecting the results of the chlamydia test.

Prepare male urine:

- The urine specimens should be centrifuged in order to collect all the particles which may contain chlamydia cells. Centrifuge the urine (at least 15 min) at 10,000 rpm for 10 minutes.
- Carefully drain the excess and add 5 drops of extraction solution A to the tube, re-suspend the pellet with a disposable pipette and incubate at room temperature for 2 minutes.
- Transfer the suspension into the extraction tube with a disposable pipette, add 5 drops of extraction solution B and mix (e.g. by means of bringing the solution up into a pipette repeatedly). The sample can now be placed in the test cassette without additional incubation time.
3. Fit the dropper tip on top of the extraction tube.

4. Add 3 drops of the extracted sample from the extraction (approx. 120µl) tube into the round test cassette sample well marked with an S.

5. Wait until pink lines appear. The test result should be read off within 15 minutes after adding the extracted suspension to the sample. Depending on the concentration of Chlamydia on the swab, some results may be visible after just 1 minute. However, to confirm negative results, the full reaction time of 15 minutes is required. After 20 minutes no further results should be interpreted.

**POSITIVE**: Two distinct colored lines appear. One line should be in the control line region (C) and another line should be in the test region (T).

**NEGATIVE**: A coloured line appears only in the control band region (C). No red band is visible in the test line region. This indicates that the sample does not contain any chlamydia antigen or that the quantity of chlamydia antigen was below the detection limit.

**INVALID**: Control line fails to appear. Insufficient specimen volume or incorrect procedural errors may cause invalid results. Review the procedure and repeat the test with a new test. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

**QUALITY CONTROL**

- The Chlamydia antigen test includes a procedural control. A pink coloured line in the control region of the membrane indicates proper performance of the test and reagents.
- Good laboratory practice includes the use of external controls to ensure proper kit performance. Two commercial controls should be carried out on each lot. The two controls should consist of a negative control and a positive control with low levels of Chlamydia. Use of the weakly positive control will ensure that the test has not been adversely affected and that Chlamydia are detected at the stated sensitivity of the test system.

**CLINICAL SIGNIFICANCE**

The genus Chlamydia is made up of three species: Chlamydia trachomatis, Chlamydia pneumoniae (1), a primarily human pathogen, and Chlamydia psittaci, a primarily animal pathogen. Chlamydia trachomatis consists of 15 known serovars. These are associated with urogenital infections and lymphogranuloma venereum (LGV). Chlamydia trachomatis infections are the most common bacterial sexually transmitted diseases. In the USA there are about 4 million new cases every year, mainly cervicitis and non-gonococcal urethritis (6). This organism also causes conjunctivitis and pneumonia in children (2, 4–7). Chlamydia trachomatis infections have both a high prevalence and asymptomatic carriage rate, with frequent serious complications in both women and neonates. Complications of chlamydiae infections in women include cervicitis, urethritis, endometritis, pelvic inflammatory disease (PID) and increased incidence of ectopic pregnancy and infertility. Vertical transmission of the disease from the mother to the neonate during parturition can result in inclusion conjunctivitis and pneumonia (8). In men at least 40% of all cases of non-gonococcal urethritis are associated with chlamydia infections and epididymitis (6). Approximately 70% of women with endocervical infections and up to 50% of men with urethral infections are symptomatic (8). Chlamydia psittaci infections are associated with respiratory diseases in individuals exposed to infected birds and are not transmitted from human to human. Chlamydia pneumoniae, which was first isolated in 1983, is associated with respiratory infections and pneumonia (2). Traditionally chlamydia infections have been diagnosed by the detection of chlamydial in tissue culture cells or polymerase chain reaction (PCR). Together with PCR, the culture method is the most sensitive and specific laboratory method, but it is labour-intensive and expensive, it takes a long time (2–3 days) and is not routinely available in most institutions (2, 3, 7). Direct tests such as immunofluorescence assay (IFA) require special equipment and a skilled operator to read the results.

**EXPECTED VALUES**

For women attending STD clinics and other high-risk populations, the prevalence of Chlamydia infection has been reported to be between 20% and 30%. In a low-risk population such as those patients attending obstetrics and gynecology clinics, the prevalence is approximately 5% or less. Reports show that for men attending STD clinics, the prevalence of Chlamydia infection is approximately 8% in asymptomatic men and 11% in symptomatic men. Normal carriage rates of Chlamydia in asymptomatic men are less than 5%.

**ANALYTICAL PERFORMANCE**

**Sensitivity**

To determine the analytical sensitivity of the chlamydia antigen test, specimens with various types of chlamydia were examined. The detection level of the chlamydia antigen test was set at 1.0 x 10^7 organisms per test.

**Specificity**

The antibody mix used in the antigen test for Chlamydia is targeted at a genus-specific epitope, which is present in all 15 Chlamydia serovars. In addition, Chlamydia psittaci and Chlamydia pneumoniae strains have been tested with the chlamydia antigen test and gave positive results. Cross reactivity with other organisms has been studied using suspensions of 10^7 CFU/ml specimen material. The following organisms were not detected using the chlamydia antigen test:

- Acinetobacter calcoaceticus
- Neisseria meningitidis
- Proteus vulgaris
- Neisseria lactamica
- Salmonella typhi
- Escherichia coli
- Chlamydia psittaci spp.
- Gardnerella vaginalis
- Staphylococcus aureus
- Streptococcus faecalis
- Candida albicans
- Streptococcus faecium
- Neisseria gonorrhoea
- Pseudomonas aeruginosa
- Neisseria gonorrhoea
- Trichomonas vaginalis

**NOTES**

1. Do not mix kit components from different lots. Do not confuse solution bottle caps.
2. Do not use test cassettes or swabs with damaged packaging.
3. Do not dismantle test cassette.
4. Use test cassette only once.
5. The materials used in the test cassette (for example antibodies) are potentially infectious. With appropriate application however they pose no danger.
6. Use appropriate precautions in the collection, handling, storage and disposal of the specimens and used kit contents. All specimens, reagents and controls should be treated as potentially infectious components.
7. When the test procedure is complete, the swabs must be disposed of in accordance with the guidelines relating to contact with potentially infectious materials.
8. Avoid cross-contaminations of the samples by using separate swabs and extraction tubes for each sample.
9. Do not eat, drink or smoke in the area where specimens and kit reagents are handled. Wear protective clothing such as laboratory coats and disposable gloves for collection and testing of specimens.
10. As with all diagnostic tests, the definitive clinical diagnosis should not be based on a single test, but should only be made by a doctor after all the clinical and laboratory results have been examined.
11. The chlamydia antigen test does not specifically differentiate between C. trachomatis, C. pneumonia or C. psittaci. Detection of chlamydia is dependent on the concentration of organisms present in the specimen. This may be affected by specimen collection methods and patient factors such as age, previous history of STDs, presence of symptoms, etc.

**REFERENCES**