GONORREA Ag Card

For in*Vitro* **diagnostic use only**

Rapid immunochromatographic test for qualitative detection of *Neisseria Gonorrhea* **from swab and human urine**

I. INTRODUCTION AND INTENDED USE

Gonorrhea Ag Card is a rapid test for the visual detection of Gonorrhea antigen, in the secretory specimen from urogenital system, as in aid in the diagnosis of gonococcus infection. Gonorrhea is one of the common sexually transmitted diseases (STD) in the whole world. Incidence of a disease is increasing year after year. It is the key approach to early and fast diagnosis for early treatment and shutting infection off. Nowadays Neisseria Gonorrhea is the major pathogenic bacteria for many developing countries.

II. PRINCIPLE

Gonorrea Ag Card is a rapid immunochromatographic test for the visual detection of Gonorrhea antigen in either secretory specimens. This test adopts double antibody sandwich method. When the specimen is added into the test device, the specimen is absorbed into the device by capillary action, mixes with the antibody-dye conjugate, and flows across the pre-coated membrane. When the Gonorrhea antigen levels are at or above the target cutoff (the detection limit of the test), Gonorrhea antigen in the specimen binds to the antibody-dye conjugate and are captured by monoclonal antibody immobilized in the Test region (T) of the device. This produces a colored Test band and indicates a positive result. When the Gonorrhea antigen levels are zero or below the target cutoff, there is not a visible colored band in the Test region (T) of the device. This indicates a negative result. A colored line will appear at the Control region (C), if the test has been performed properly.

III. COMPOSITION OF KIT

Each kit contains:

1. **Neisseria Gonorrhea Card** (**25 items**): the cards are stored in a sealed pouch.
2. **Extracton Solution A (1x7.5 mL)** in plastic dropper bottles containing NaOH, pH 11
3. **Extracton Solution B (1x2.5 mL)** in plastic dropper bottles containing HCl, pH 3
4. **Positive Control (1 x 0.5 mL).:** N.1 vial with dropper containing non-infectious components, sodium azide (NaN_3) as preservative
   Negative Control: use Extraction Solution : **Solution A (3 drops) + Solution B (1 drop)**
5. **Tampone swab (25 items)**
6. **Vial with dropper (25 items)**
7. **Instructions leaflet (1 item)**

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. Gonorrea Ag 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, handing, storage and disposal of specimens 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 pretreating 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.
- As with all diagnostic tests, a decisive clinical diagnosis should not be based on the result of a single test, but should only be made by a physician after all clinical and laboratory findings have been evaluated.
- 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.

VI. SPECIMEN COLLECTION AND PREPARATION

For the best performance of any Gonorrhea Ag Card, 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. Use the swab provided with the kit. Alternatively any shafted swabs with rayon, cotton or dacron tips may be used. Insert 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 *Neisseria Gonorrhea* 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.
5. 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) Urine Specimens:

Collect 15-30 mL of clean first morning urine in a sterile urine cup. First morning urine specimens are preferred to achieve the highest concentrations of *Neisseria Gonorrhea*. Although urine specimen is workable, the swab specimen is more recommended for higher sensitivity.

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 (4-8°C). The urine specimens can be stored refrigerated (4-8°C). Do not freeze. All specimens should be allowed to reach a room temperature of 10-30°C before testing.

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 or Urethral swab specimens:

- 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 5 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 2 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.
- The extracted specimen can remain at room temperature for 60 minutes without affecting the result of the test.

Preparation of urine specimens:

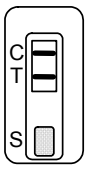
- Mix the urine specimen by inverting the container. Transfer 15 mL of the urine specimen into a centrifuge tube, centrifuge at 3,000 rpm for 15-20 minutes.
- Carefully pour odd the supernatant and discard. Keep the tube inverted and remove any supernatant from the rim of the tube by blotting onto absorbent paper. If necessary, can be precipitated and then centrifuge as in the above step.
- Add 6 drops of Extraction Solution A to the centrifugal tube vertically, vigorously mix (when necessary, vortex mixer can be used to vortex and mix) until the suspension is homogeneous. Transfer the suspension into the extraction tube, and **let it stand for 2 minutes**. Then add 2 drop of Extraction Solution B to the extraction tube vertically, vortex or tap the bottom of the tube to mix the solution**.**

B) TEST PROCEDURE

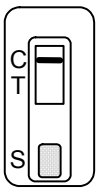
- Follow instructions for specimen collection and extraction
- Remove Gonorrea Ag Card from its protective pouch and place on a level surface, clean and dry. Label the device with patient's name or control number.
- Place the cap on the extraction tube. Add 3 drops (approx. 150 µL) of extracted sample from Extraction tube to the sample well.
- Wait for test band (S) to appear. The test results should be read in 10-15 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. **Do not interpret result after 15 minutes.**

PROCEDURE FOR CONTROLS
Add 3 drops (150µL) of **Positive/Negative control** on the sample well; read after 10 minutes.

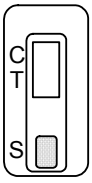
VIII. READING TEST RESULTS



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. PERFORMANCES CHARACTERISTICS

A) Detection limit
For Neisseria gonorrheae, the lowest detection limit of the test is 1×10^5 CFU/ml.
B) Sensitivity and Specificity
200 patients who have the urogenital discomfort syndromes were tested for gonorrheal infection and their swab specimens were tested
in parallel using this Gonorrea Ag Card and a conventional culture test. For calculating sensitivity and specificity, the cell culture result is assumed to be 100% accurate. Of the 200 samples tested, 32 were culture positive and 168 were culture negative.
The results are shown as follow:
Sensitivity of Gonorrea Ag Card: 30/32×100%=**93.8%**
Specificity of Gonorrea Ag Card: 164/168×100%=**96.7%**
C) Cross-reactivity
Cross reactivity with other organisms has been studied using suspensions of 10^5-10^8 CFU/ml. The following organisms were not detected using Gonorrea Ag Card: *Satin Monilia, Herpes simplex virus, Neisseria, Pseudomonas aeruginosa, Streptococcus, Mycoplasma hominis, Candida albicans Staphylococcus aureus, Escherichia coli and Trichomonas vaginalis.*
D) Precision
1. Within run precision was determined by using 10 replicates of four different specimens containing different concentrations of antigen. The negative and positive values were correctly identified 100% of the time.
2. Between run precision was determined by using the four different specimens containing different concentrations of antigen in 3 different lots of test devices. Again negative and positive results were correctly identified 100% of the time.

X. LIMITATIONS

1. The Gonorrea Ag Card is for in vitro diagnostic use only. The test should be used for the detection of *Neisseria Gonorrhea* only and for specimens collected from the endocervical or urethral regions.
2. Specimen with an excessive amount of mucus or blood may give false positive results.
3. Like all immunological tests for *Neisseria Gonorrhea*, this test cannot distinguish between biologically active and inactive organisms.
4. This test will only indicate the presence or absence of *Neisseria Gonorrhea* in the specimens and should not be used as the only basis for the diagnosis of *Neisseria Gonorrhea*.
6. If clinical symptoms persist ant 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 on a synopsis of laboratory results, clinical investigations and patient's history.

XII. BIBLIOGRAPHY (see Italian version)

CONTENTS (25 test) **REF VQ81602**

1.	Gonorrea Ag Card	25 items
2.	Extraction solution A in plastic dropper bottles	1 x 7.5 mL
3.	Extraction solution B in plastic dropper bottles	1 x 2.5 mL
4.	Positive Control	1 x 0.5 mL
5.	Sterile Swabs	25 items
6.	Vial with dropper	25 items
7.	Instructions leaflet	1 item

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

EDMA CODE 15 01 90 00 CE IVD

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