TRICHOMONAS VAGINALIS

For in *Vitro* diagnostic use only

Diagnostic rapid test for the detection of *Trichomonas vaginalis* in urogenital swabs and urine

I. INTRODUCTION AND INTENDED USE

Trichomoniasis is most commonly transmitted through unprotected sexual intercourse. The vagina is the most common site of infection in women while the urethra is the most common site of infection in men. Trichomoniasis accounts for 15-20% of the cases of vaginitis. It occurs in both men and women and is caused by an infection with the single-celled parasite *Trichomonas vaginalis*. Infection with *Trichomonas vaginalis* is frequently associated with other sexually transmitted diseases and helps spreading the **HIV** virus.

II. PRINCIPLE OF THE TEST

The test strip in the device includes: 1) a pink-colored conjugate pad containing colloidal gold coupled with *Trichomonas vaginalis* monoclonal antibodies, and 2) a nitrocellulose membrane containing a test line (T-line) and a control line (C-line). The T-line is coated with *Trichomonas vaginalis* antibody, and the C-line is coated with antibodies anti-species. When *Trichomonas vaginalis* antigens are present in the specimen, the T-line will become a pink-colored band. If antigen to *Trichomonas vaginalis* are not present or are present below the detectable level, no T-line will develop. The C-line should always appear as a pink-colored band regardless of the presence of antigen to *Trichomonas vaginalis.* The C-line serves as an internal qualitative control of the test system to indicate that an adequate volume of specimen has been applied and the flow occurred.

III. REAGENTS AND MATERIALS

Each kit contains:

1. Trichomonas Vaginalis strip (20): each sealed with a test strip and a dessicant in the pouch.
2. Extraction buffer-Diluent (1 x 20 mL): Dilution buffer, containing NaN_3 (<0,1%), a detergent, and proteins.
3. 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 buffer-Diluent
4. Instruction for use (1)

Required materials (not supplied)

Test tubes 3 or 5 mL - Specimen collection container - Disposable gloves – Sterile swabs

IV. STORAGE AND STABILITY

Store as packaged in the sealed pouch either at refrigerated or room temperature (+2ºC - +30ºC). Do not freeze.

V. SPECIAL PRECAUTIONS

1) The kit is for professional use and for in vitro diagnosis only.
2) Do not use after expiration date. Do not use the test if pouch is damaged.
3) Read carefully the Instruction for use before using this kit.
4) The test should remain in the sealed pouch until use.
5) All operations linked to the use of the test must be performed in accordance with Good Laboratory Practices.
6) All the specimens should be considered potentially hazardous and handled in the same manner as an infectious agent.
7) The test should be discarded in a proper biohazard container after testing.
8) The test must be carried out within 2 hours of opening the sealed bag.

VI. SPECIMENS COLLECTION

URINE SAMPLE

For men, the urine sample be performed to test Trichomonas V infection. as it usually contains the high secreted protein of Trichomonas V parasite. Collect a urine specimen in a clean glass, plastic, or wax coated container. Do not use preservatives. If the test is not run immediately following collection of the sample specimen, but is to be run within 24 hours following collection. Please take the equal volume of sample buffer of kit into test tube, and mix well, the diluent sample should be refrigerated (2-8°C), and brought back to room temperature (15-28°C) before testing. If testing is delayed more than forty-eight hours, the specimen should be frozen at -20°C or lower. Prior to testing, the frozen specimen must be completely thawed, thoroughly mixed, and brought to room temperature.

SWAB SAMPLE

Collect specimen with a sterile swab from vaginal cavity, or Glans. Process the swab as soon as possible after collection specimen. This test does not require live organisms for processing. If you cannot dry the swabs or cannot perform test immediately, extract the swabs in Sample Buffer as per protocol and store the aqueous extracted specimen at 2–8°C for up to 24 hours. Alternatively, dried swabs can be stored at 2-8°C for up to 24 hours prior to extraction and testing.

VII. PROCEDURES

Allow the tests, samples and buffers to reach to room temperature (15-30ºC/59-86ºF) prior to testing. Do not open the pack until ready to perform the assay.

For Urine Sediment

1. Collect to 10-15ml of Urine into clear centrifuge tube.
2. Centrifuge 3000 rpm for 5 minutes.
3. Discard to supernatant.
4. Use Votex or mixer to mix bottom liquid of centrifuge tube for 3-5 seconds.
5. Pipette to 0.5ml of sample buffer into centrifuge tube and mix well with sediment in 5-10 seconds
6. Holding the strip vertically, place it in the specimen test tube. Do not immerse the strip past the maximum line.
7. The test strip can be left in the tube or removed from the tube when red dye begins to migrate through the test line.
8. Read the test results at 15 minutes.
9. Please read the test card in 15 minutes if the reading time be after in 30 minutes the test will be considered as a failure.
10. Interpretation of Result by eyes.

For Urine sample
It is recommended to collect the first urine of the day for higher accuracy
1. Pipette to 0.5ml of sample buffer into test tube.
2. Pipette to 0.5ml of urine into sample tube with buffer, the value be made to equal volume to mix.
3. Mix well in 5-10 seconds.
4. Remove a Trichomonas V Test Strip from its foil pouch.
5. Holding the strip vertically, place it in the specimen test tube. Do not immerse the strip past the maximum line.
6. The test strip can be left in the tube or removed from the tube when red dye begins to migrate through the test line.
7. Read the test results at 15 minutes.
8. Please read the test card in 15 minutes if the reading time be after in 30 minutes the test will be considered as a failure.
9. Interpretation of Result by eyes.

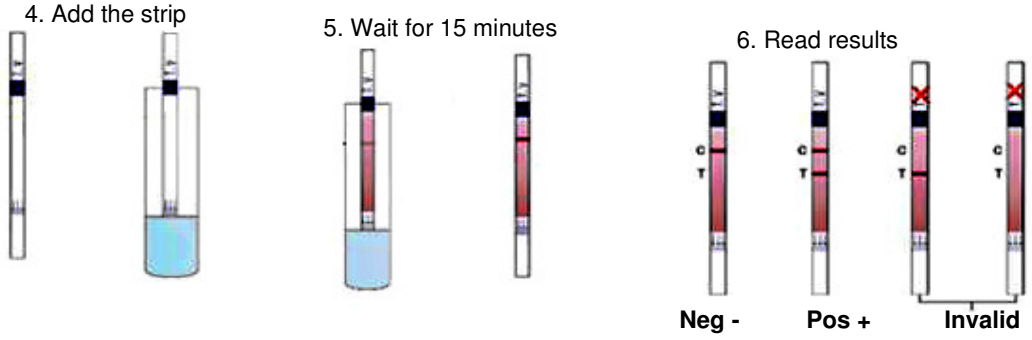
For Swab sample
1. Dispense 0.8-1.0 mL (16 drops) of Diluent into a test tube.
2. Place the sample swab into the tube. Mix the sample buffer by rotating the swab vigorously in the tube.
3. Allow the swab to soak in sample buffer for 30-60 seconds.
4. Mix again and remove the swab, pressing against the side of the tube to extract as much liquid as possible.
5. Discard the swab.
6. Remove a Trichomonas Vaginalis test from its foil pouch.
7. Holding the strip vertically, place it in the specimen test tube. Do not immerse the strip past the maximum line.
8. The test strip can be left in the tube or removed from the tube when red dye begins to migrate through the test line.
9. Read results at 15 minutes (positive results may be seen earlier)

For the Positive and Negative controls add 6 drops to an empty tube and use the same assay procedure (from point 6 on).

VIII. INTERPRETING THE RESULTS
Positive: two red lines (C) and (T) are visible in the control and test areas of the window. The intensity of the band colour in the test region is proportional to the antigen concentration in the sample. However, neither the quantitative value, nor the rate of increase in antigens can be determined by this qualitative test.
Negative: In the reading window only 1 red band appears in the control region "C". This is the control line assuring the correctness of test performance.
Invalid: No band appears in the control region. A sample should never be identified as positive if you do not see a control line. If the control line is not formed, the test is invalid and must be repeated.

1. Add the buffer
2. Collect sample
3. Mix for 30-60 seconds

4. Add the strip
5. Wait for 15 minutes
6. Read results



IX. PERFORMANCE
A. Correlation, Sensitivity-Specificity
The gold standard was the patient positive to speculum and vaginal examinations to be diagnosis Trichomonas Vaginalis infection. The total 204 cases was compared to wet mount microscopy pouch culture and Mascia Brunelli Trichomonas vaginalis Test Kit. The results are summarized as below table: the Mascia Brunelli Trichomonas vaginalis Test kit will detect soluble antigen present in vaginal samples with at least 20 organisms.

Method (N=204)	Sensitivity	Specificity
Microscopy	72.5%	78.5%
Culture	91.2%	92.8%
Trichomonas Vaginalis kit (Mascia Brunelli)	100%	99%

B. Precision
Due to the nature of the test system, precision was evaluated by testing various samples on consecutive days, by multiple clinicians at different geographic locations. Trichomonas Vaginalis Mascia Brunelli precision studies were run on artificially generated laboratory samples reflecting the known range of T. vaginalis parasite burden levels in female patients. Given the design of the product, the Trichomonas Vaginalis Mascia Brunelli performed according to expectations: the Trichomonas Vaginalis kit performed with 100% precision over the intra-assay (between day, between clinician, and between clinical site) parameter.

X. LIMITS OF THE KIT
The results obtained with this kit must be interpreted together with other clinical information and laboratory findings available to the physician.
A positive result does not preclude the possibility of infections by other pathogens.
This test provides a presumptive diagnosis of *Trichomonas V.* infections during the acute phase of infections. Samples collected after acute phases of infection may contain titles of antigens below the threshold of sensitivity of the reagent. If the test result is negative and clinical symptoms persist, additional testing using other diagnostic methods is recommended.

XI. REFERENCES see Italian version

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

CONTENT (20test)	**REF. VQ81604**
1. Trichomonas Vaginalis strip	20 strips
2. Extraction Buffer-Diluent	1 x 20.0 mL
3. Positive Control	1 x 0.5 mL
4. Instruction for use	1 item

EDMA (EDMS) CODE 1470019000 CE IVD