

TEST CARDS FOR RAPID DETECTION OF CANDIDA ANTIGEN IN VAGINAL SWAB SPECIMENS OF SYMPTOMATIC PATIENTS

Carefully read the instructions for use before testing. Perform the test in a well illuminated place.

INTRODUCTION

Candida albicans is a yeast frequently present on healthy mucous membranes of the vagina.^{1,2} However, conditions which disturb the balance of normal vaginal flora allow opportunistic growth of *Candida albicans*, which is characterized mainly by itching and the presence of a white, curd-like vaginal discharge. Redness and swelling of the epithelial surface of the vulva may also be present. *Candida* has been associated with nearly one third of all cases of infectious vaginitis.³

In the majority of cases, certain predisposing factors may contribute to creating a suitable environment for vulvovaginal candidiasis. Among these factors are: pregnancy, diabetes, obesity, malignancy, endocrine disorders, antibiotic therapy, treatment with immunosuppressive drugs, and physical barriers which inhibit normal vaginal drainage.^{1,2,3,4} Although *Candida* has been implicated as a sexually transmitted agent, this has not yet been proven.³ Some individuals appear to be more prone to recurrent *Candida* infections for reasons which are not well-defined.

Investigations have revealed that approximately 30 % of all healthy women have asymptomatic, subclinical *Candida* colonization in the vaginal tract.^{4,5} The presence of *Candida* as detected by traditional test methods does not therefore necessarily imply disease.

Common laboratory procedures utilized in the diagnosis of vulvovaginal candidiasis are direct microscopic examination of vaginal smear and yeast culturing. Both have their shortcomings from the point of view of providing the physician with accurate information on which to base treatment. Direct microscopic examination of the vaginal discharge from symptomatic patients reveals yeast forms in only fifty percent of the cases, where as positive culture isolation may indicate a subclinical infection not diagnostic of vulvovaginal candidiasis.³

Biocard™ Candida is a rapid agglutination test for the detection of *Candida* antigen directly from vaginal swab specimen. The detection level of Biocard™ Candida corresponds to the levels normally associated with clinically significant vulvovaginal candidiasis. Levels of *Candida* commonly found in the asymptomatic vaginal carriage do not generally yield a positive result.

TEST PRINCIPLE

In the Biocard™ Candida, latex particles are coated with antibodies against *Candida*. The reagent is dispensed as test doses and dried on the test card. The test is performed by adding one drop of diluted vaginal swab specimen onto the reagent dot. After mixing, the test is rotated to facilitate the formation of antigen-antibody complexes. If *Candida* antigen is present in sample it reacts with the antibody-coated test latex particles, and visible agglutination or clumping will occur. If the sample is free of *Candida* antigen the reaction mixture remains homogeneous throughout the test. The control latex reveals those infrequent samples, which spontaneously react with protein-coated latex particles.

MATERIALS PROVIDED

Biocard™ Candida 32 Tests.
Cat. no. 3-007-000

Test Cards: 8 pcs in foil pouches. Each card contains 4 test circles with dried test latex and 4 control circles with dried control latex.

Positive Control Reagent: 0,5 ml of inactivated, diluted yeast antigen.

Elution Buffer: 15 ml Sample Elution Buffer pH 7.4.

Elution Tubes: 32 pcs

Stirrer Pipettes: 32 pcs

Sterile Sampling Swabs: 32 pcs

Working Station: 1 pc

Instructions for Use: 1 pc

Biocard™ Candida 4 Tests.
Cat. no. 3-007-004

Test Cards: 1 pc in foil pouch. Card contains 4 test circles with dried test latex and 4 control circles with dried control latex.

Positive Control Reagent: 0,5 ml of inactivated, diluted yeast antigen.

Elution Buffer: 5 ml Sample Elution Buffer pH 7.4.

Elution Tubes: 4 pcs

Stirrer Pipettes: 4 pcs

Sterile Sampling Swabs: 1 pc

Working Station: 1 pc

Instructions for Use: 1 pc

Materials required but not provided: timer

STABILITY AND STORAGE

Biocard™ Candida test cards, positive control reagent and elution buffer should be stored at room temperature +2°...+27°C. The shelf life of the kit is 24 months from the date of manufacturing. Expiry date is indicated on the outer box, reagent labels and on the foil pouches containing the test cards.

Opened test card pouches should be closed without delay in a zip log bag, or resealed for example with masking tape to avoid moisture absorbency. If possible the opened pouches should be stored in an excicator. Do not remove the drying agent from the foil pouch.

PRECAUTIONS AND LIMITATIONS

- Biocard™ Candida test should be used only for *in vitro* detection of *Candida* antigen according to the instructions for use. If the instructions for use are not followed in detail, the outcome of the test may be false.
- Do not use product after the indicated expiry date
- Infectious agents may be present in the test specimen. Therefore, all specimens should be regarded and handled as a potential biohazard.
- Although the reagents do not contain infectious agents of *Candida* they are prepared from biological materials and should be regarded and handled as a potential biohazard.
- General laboratory procedures and precautions should be followed in the handling and disposal of specimens and testing materials.

- Reagents contain 0,09% sodium azide. Positive control contains formaldehyde.
- Do not mix reagents from kits with different lot numbers.
- Do not use the test cards if the foil pouch is damaged.
- Do not use damaged accessories
- Take care not to touch the latex reagents dried onto the test card, as moisture from fingertips can displace the reagents and negatively affect the results.
- The swab containing the vaginal specimen must not come in contact with 10% KOH
- Do not use cards with a latex reagent spot which sticks to the foil wrapper or is difficult to re-suspend with visible granulation in either the test or control latex.
- Do not use expired reagents or test cards not showing proper performance with the positive and negative control reagents.
- Use only swabs provided with the kit.
- Do not re-use.
- A diagnosis should not be made solely according to the Biocard™ Candida-test result. The result should be used in conjunction with additional diagnostic information available for the physician.

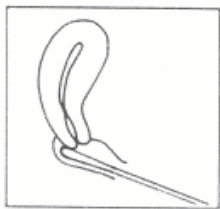
SAMPLES

Using one of the swabs provided with the kit take a deep vaginal swab. Insert the swab into one of the test tubes provided. Dispense approximately 10 drops (app. 400 µl) of Biocard™ buffer into the tube so that the head of the swab is immersed in the buffer in the bottom of the test tube. Agitate the head of the swab in the buffer for 10 seconds. Finally rotate and squeeze the head of the swab against the tube wall to recover as much fluid from the swab as possible. Discard the swab. (NOTE: the swab must not come in direct contact with 10% KOH. If a 10% KOH slide is going to be prepared from a specimen swab prior to testing with Biocard™ Candida, it is recommended the swab be rolled onto glass slide and then set aside for subsequent Biocard™ Candida testing. The 10 % KOH may then be added and mixed with the specimen already transferred on the slide to create a suspension for microscopic analysis. The presence of 10% KOH in the Biocard™ Candida test system will alter immunological reaction and cause erroneous results.)

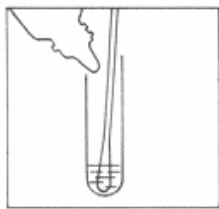
The sample solution can be tested immediately or within 4 hours. Alternatively, the sample solution can be stored in a refrigerator for up to 48 hours before testing.

Avoid sampling of mucus and blood. If mucus matter is present in excess it will disturb the test reaction by causing a false positive test and control latex reaction. The interpretation of the test result will become difficult if the specimen contains red blood cells in excess.

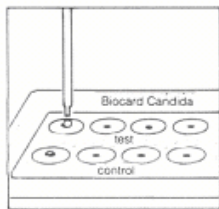
If the test can not be performed immediately after taking the sample, the sampling stick containing the sample may be stored in a clean, dry vial for up to 72 hours.



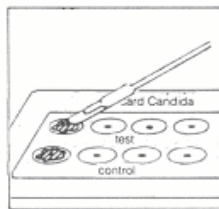
Take a deep vaginal specimen



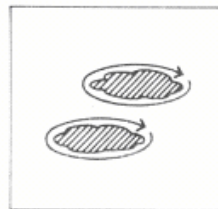
Eluate the specimen in the Biocard Candida Buffer



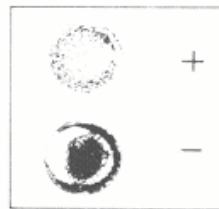
Pipette one drop of the eluate on a control latex dot and one drop on a test latex dot



Wait for 10 - 20 sec. Mix and spread the suspensions in the control and test circles



Smoothly tilt and rock the test card so that the suspensions slowly rotate in the circles and examine for signs of agglutination



A definite blue-coloured agglutination in the test circle and the absence of agglutination in the control circle indicates the presence of *Candida* antigen

TEST PROCEDURE

1. Remove any stored specimen elutes from the refrigerator and allow them to reach room temperature. This will take from 5 to 15 minutes.
Open Biocard™ *Candida* test package. Take the number of test cards required out of the test package. If desired, cut the number of tests needed from the test card. Close the opened pouch in the zip lock bag without delay.
2. Mix the eluted sample thoroughly. Using a disposable stirrer-pipette pipette one free-falling drop (30 µl) of the sample elute first to one of the test latex dots. (While pipetting, hold the pipette in a vertical position and avoid formation of bubbles on the test card.) Wait 10 – 20 seconds. Mix and spread the sample with the stirrer-pipette, first in the control circle and then in the test circle, to make two homogeneous blue suspensions. Start the timer.
3. Smoothly tilt and rock the test card so that the suspensions slowly rotate in their circles and examine for signs of agglutination.
4. A positive reaction (*Candida* antigen is present in the specimen) is indicated by a definite blue-coloured agglutination in the test latex circle and the absence of definite agglutination in the control latex circle within 2 minutes reaction time.

A negative reaction is indicated by the absence of definite blue-coloured agglutination in the test latex circle within 2 minutes reaction time.

If the test latex reaction is positive but agglutination is observed also in the control latex circle, the specimen includes components which aggregate protein-coated latex beads and the test result cannot be interpreted. In this case it is recommended to repeat the test with a new specimen or to test the specimen with a culture method.

NOTE

A positive reaction can be read as soon as it appears. If the *Candida* colonization is very heavy the agglutination may occur during the mixing of the elute and the test latex reagent. If the sample elute contains solid particles be careful not to confuse these with agglutination.

CONTROLS

Proper performance of Biocard™ *Candida* can be tested using Biocard™ *Candida* Positive Control Reagent as a positive control and Biocard™ *Candida* Sample Elution Buffer as a negative control.

Mix the control reagent by inverting the vial. Dispense one free-falling drop (30 µl) onto the reagent dot in the control circle and one drop onto the reagent dot in the test circle. Wait for 10 – 20 seconds. Mix and spread the suspension with the stirrer-pipette, first in the control circle and then in the test circle to make two homogeneous blue-coloured suspensions. Start the timer. Continue as described in section 3 of the test procedure.

Biocard™ *Candida* Positive Control Reagent should always give a positive reaction in the test latex circle and no reaction in the control latex circle when tested as described above.

Perform a negative control test as described above but replacing the positive control reagent with Biocard™ *Candida* sample elution buffer.

The control test performed with neat elution buffer should give no reactions in both the test and the control latex circles.

SENSITIVITY AND SPECIFICITY

The detection level of Biocard *Candida* corresponds to the levels normally associated with clinically significant vulvovaginal candidiasis. Levels of *Candida* commonly found in the asymptomatic vaginal carriage do not generally yield a positive result. Absolute sensitivity of the Biocard *Candida* has been determined to be 10⁶ CFU.

The Biocard *Candida* has been tested against the following strains of *Candida* and gave positive results with all tested strains listed below.

Candida albicans ATCC 3153
Candida albicans ATCC 36802
Candida albicans ATCC 28367
Candida albicans ATCC 24433
Candida albicans ATCC 90028
Candida albicans serotype A
Candida guilliermondii ATCC 6260
Candida krusei ATCC 14243
Candida glabrata ATCC 66032
Candida tropicalis ATCC 9968

The Biocard *Candida* has been tested against the following bacterial strains and gave negative results with all tested strains listed below.

Staphylococcus aureus ATCC 25923
Staphylococcus aureus strain 510
Staphylococcus aureus strain 530
Cryptococcus neoformans
Group B streptococcus ATCC 123 86
Streptococcus faecalis ATCC 19433
Streptococcus agalactiae strain 59
Staphylococcus epidermis
Staphylococcus saprophyticus strain 2866
Acinetobacter strain 9202
Neisserie gonorrhoeae strain 717
Trichomonas vaginalis
E.coli
Bacteroides fragilis
Chlamydia
Lactobacillus sp.
Gardnerella vaginalis
Herpes simplex virus

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