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|---|---|---|---|---|
| IVD | See external label | Σ=25 tests | 2°C-28°C | REF Cat # 176522-1 |

**OneStep**

# Mononucleosis

**RapiCard™ InstaTest**

**Cat. # 176522-1**

## INTENDED USE

The Cortez Diagnostics Inc. OneStep Mono RapiCard™ InstaTest is a rapid, convenient method for detecting infectious mononucleosis heterophile antibodies at the early stage of infection. With the combination of a mononucleosis antigen isolated from bovine red cell membranes and a mouse antibody against human IgM (μ), the antibodies in a serum or plasma sample are detected in a little over five minutes with a high degree of sensitivity.

## SUMMARY AND EXPLANATION OF THE TEST

Infectious mononucleosis (IM) is an acute, benign, communicable disease most commonly caused by the Epstein-Barr virus (EBV) and characterized by lymphadenopathy, pharyngitis, fever, atypical lymphocytosis, and splenomegaly.[1]

The diagnosis of IM is usually based on evaluation of characteristic clinical, hematologic, and serologic changes. Serologic diagnosis of IM is demonstrated by the presence of heterophile and EBV antibodies in the sera of patients.[2] The heterophile antibodies belong to the immunoglobulin M (IgM) class.[3]

The Cortez OneStep Mono RapiCard™ InstaTest is an immunochromatographic assay which utilizes the heterophile antigen isolated from bovine red cell membranes, and a mouse monoclonal antibody, to selectively identify the IM heterophile antibodies in serum with a high degree of sensitivity.[4]

## PRINCIPLE OF THE TEST

The Cortez OneStep Mono RapiCard™ InstaTest includes an immunochromatographic device in which a dye-conjugated monoclonal antibody against human IgM, and a heterophile antigen immobilized on the membrane are used to produce a distinctive visual pattern registering the presence of IM heterophile antibodies in the test sample.

In the test procedure, patient serum or plasma is pretreated with an extraction buffer and then added to the sample

well "S" of the test device with the aid of a transfer pipette. Labeled antibody-dye conjugate binds to the IM heterophile antibodies (if the latter are present in the specimen) forming a colored dye complex. This complex is captured by the heterophile antigen immobilized in the test zone "T" of the membrane, and is visible as a pink-rose color test band. A colored conjugate is captured in the control zone "C" of the membrane by an immobilized reagent, where it is visible as a pink-rose control band.

A pink-rose band in the test zone with the control band visible is considered a positive result, and indicates that the patient specimen contains IM antibodies. The presence of the control band indicates that the test device is functioning correctly, and that the test reagents and sample specimen have properly mixed and migrated through the test membrane.

## REAGENTS AND MATERIALS PROVIDED

1. Testing Device, 25 pcs.
2. Test Buffer, 5.0 ml
3. Transfer Pipette sealed inside each pouch with the Testing Device.
4. Test Instructions

## MATERIALS REQUIRED BUT NOT PROVIDED

1. Specimen collection container.
2. Clock or timer.
3. Centrifuge capable of 1,000 *g* centrifugal force (for centrifugation of whole blood specimens).

## STORAGE AND STABILITY

Store the test kit below 28°C; do not freeze. Refer to the expiration date for product stability.

## WARNINGS AND PRECATIONS

1. Preclude any pipetting by mouth.
2. Do not allow smoking or eating in areas where specimens are handled and tested.
3. Wear disposable gloves while handling kit components or specimens. Wash hands thoroughly afterwards.
4. Avoid splashing or aerosol formation.
5. Clean up spills thoroughly using an appropriate intermediate-to-high level disinfectant.
6. Decontaminate and dispose of all specimens and potentially contaminated materials as if they were infectious.
7. Do not use the reagents or test devices after the expiration date.
8. For *in vitro* diagnostic use only.

## SAMPLE COLLECTION AND PREPARATION

Collect blood aseptically by venipuncture into a clean tube without anticoagulants. Permit blood to clot for twenty to thirty minutes at room temperature. Centrifuge to obtain clear serum and transfer serum into a clean plastic or glass tube. The test may be performed using human serum or plasma.

If specimens are not immediately tested they should be refrigerated at 2-8º C. For storage periods greater than three days, freezing (-20ºC) is recommended. Avoid repeated freezing and thawing. If specimens are to be shipped, they should be packed in compliance with federal regulations covering the transportation of etiologic agents. Specimens containing precipitate may yield inconsistent test results. Such specimens must be clarified prior to assaying. 0.1% or less sodium azide will not affect the test results.

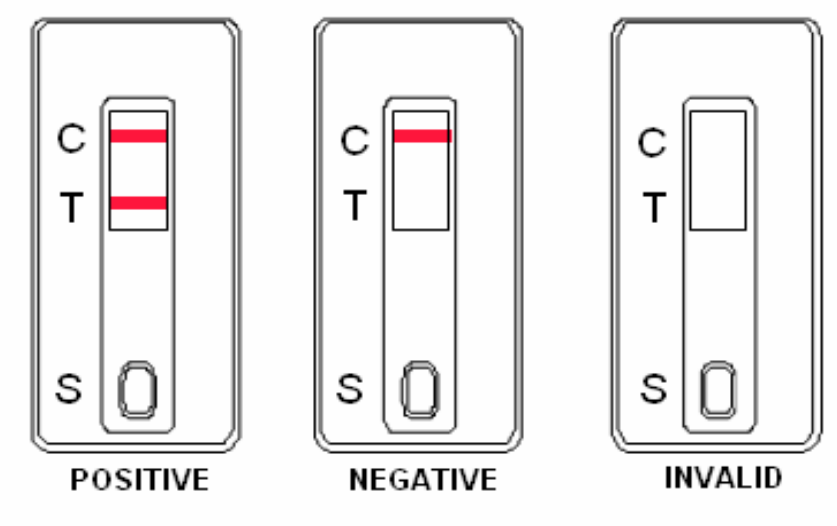
## TEST PROCEDURE

*NOTE: Several tests may be run together; however a different Transfer Pipette and Testing Device must be used for each specimen. Bring the kit components to room temperature before performing the test.*

## PERFORMING THE TEST

1. Open a foil pouch by tearing along the "notch". Remove the Testing Device and Transfer Pipette. Place the Testing Device on a level surface with windows facing up.
2. Using the Transfer Pipette, add 1 drop of the sample (serum or plasma) into the sample well "S' of the Testing Device.
3. Using the vial dropper tip, add 3 drops of Test Buffer into the sample well "S".
4. Read the results at five minutes.
   *IMPORTANT: To avoid an incorrect reading or an invalid result, do not interpret test results after more than five minutes.*

## INTERPRETATION OF RESULTS




1. **Positive .** In addition to the line in the control zone (C) of the membrane, a rose-pink line is visible in the test zone (T). The sample should be considered IM positive.
2. **Negative.** One colored line appears in the control zone with no apparent band in the test zone. The sample may be considered IM negative.
3. **Invalid.** If there are no pink-rose bands visible in both the control and test zones, or a pink-rose band is visible only in the test zone and not in the control zone, then the test is invalid. It is recommended that the specimen be retested.

## LIMITATIONS OF THE TEST

1. The Cortez OneStep Mono RapiCard™ InstaTest is limited to the detection of IM heterophile antibodies in serum, plasma, or recalcified plasma.
2. The test is for *in vitro* diagnostic use only.
3. Although the Cortez OneStep Mono RapiCard ™ InstaTest is very accurate in detecting IM, a low incidence of false results can occur.
4. If inconsistent or questionable results are obtained, and EBV infection is suspected, the test should be repeated using a fresh specimen.

5. As with all diagnostic tests, a definitive clinical diagnosis should not be based on the results of a single test, but should only be made by the physician after all clinical and laboratory findings have been evaluated.

## INSTRUCTIONS

Semi-Quantitative Procedure

**Step 1**: Make serial dilutions of each specimen using saline (0.9% sodium chloride) and 12x75mm test tubes as described in the chart below. Mix each tube before making the next dilution.

| Tube # | Specimen Dilution | Add 12 x 75mm tube: |
|---|---|---|
| 1 | Undiluted Specimen | - - |
| 2 | 1:2 | 0.5 ml patient specimen + 0.5ml saline, mix |
| 3 | 1:4 | 0.5 ml of Tube 2+ 0.5ml saline, mix |
| 4 | 1:8 | 0.5 ml of Tube 3 + 0.5ml saline, mix |
| 5 | 1:16 | 0.5 ml of Tube 4 + 0.5ml saline, mix |
| 6 | 1:32 | 0.5 ml of Tube 5 + 0.5ml saline, mix |
| 7 | 1:64 | 0.5 ml of Tube 6 + 0.5ml saline, mix |
| 8 | 1:128 | 0.5 ml of Tube 7 + 0.5ml saline, mix |
| 9 | 1:256 | 0.5 ml of Tube 8 + 0.5ml saline, mix |

**Step 2**: Proceed to test the tube #1 through 9 according to the "Qualitative Procedure"

## INTERPRETATION OF RESULTS

The IM titer defined as the reciprocal of the highest dilution which gives a positive result. If positive results are observed in all dilutions through tube #9, then addi- tional serial dilutions from tube #9 are to be performed until a negative result is obtained. As the heterophile antibody titer increases a corresponding increase in antibody concentration is observed.

**Example Data:** **Cortez Mono**

| Tube # | Specimen Dilution | Initial Specimen | 2nd Specimen |
|---|---|---|---|
| 1 | Undiluted Specimen | Positive | Positive |
| 2 | 1:2 | Positive | Positive |
| 3 | 1:4 | Positive | Positive |
| 4 | 1:8 | Positive | Positive |
| 5 | 1:16 | Negative | Positive |
| 6 | 1:32 | Negative | Positive |
| 7 | 1:64 | Negative | Positive |
| 8 | 1:128 | Negative | Negative |
| 9 | 1:256 | Negative | Negative |

In the above Example Data, specimen dilutions in tubes #5 thru 9 give negative results but tube #4 is positive therefore the titer should be reported as less than 16. If sequential titer determinations are done and the initial specimen (refer to example data) produces a lower antibody titer than the second specimen, it is indicative that the antibody level is rising and that the disease is progressing. Conversely, if the second specimen produces a lower titer, it is indicative that the antibody level is falling and that the disease is convalescing.

**Note:** Antibody titers obtained with Cortez OneStep Mono must not be compared to titers obtained with kits from competitors since there are differences in assay sensitivity.

## PERFORMANCE CHARACTERISTICS

1. **Specificity and Sensitivity** : The Cortez OneStep Mono RapiCard™ InstaTest was evaluated and compared with a hemagglutination slide test and a latex particle agglutination slide test for heterophile antibody and was found to be sustantially equivalent. A total of 145 serums which were collected from outpatient clinical laboratories were tested. Off these 90 were confirmed heterophile antibody negative, 55 were confirmed heterophile antibody positive using the traditional latex particle agglutination. Three samples tested negative with traditional latex particle agglutination and positive with Cortez OneStep. Two samples tested positive with traditional latex particle agglutination and negative with Cortez OneStep. The results of this comparison evaluation are shown in the table below.

| | Positive | Negative |
|---|---|---|
| Latex Particle Positive (55) | 53 | 2 |
| Agglutination Negative (90) | 3 | 87 |

Compared with a conventional Latex particle agglutination for the detection of antbodies to heterophile antigen from serum specimens. Cortez OneStep Mono RapiCard™ InstaTest demonstrated a sensitivity of 96.3% (55-2/55) and a specificity of 96.6% (90-3/90).

2. **Precision**:

Intra-Assay: Within-run precision was determined using the same 3 specimens containing negative, weak positive and positive values. The negative, weak positive values were correctly identified 100% of the time.

Inter-Assay: Between-run precision was determined using the same 3 specimens of negative, weak positive, and positive control of heterophile antibody in 11 dependent assays with three different lots of reagents. Again, the negative, weak positive and positive findings were correct 100% of the time.

3. **Cross-Reactivity**: The performance reactivity of the test for detecting infectious mononucleosis heterophile antibodies has been shown to be negative by utilizing the serum specimen containing HBsAb, antibody to HCV, IgM anti- body to Hepatitis A, antibody to H. pylori, antibody IgM to rubella IgM, antibody to CMV, antibody IgM to TOXO.

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