Chemtrue®

Mono Rapid Test

For *in vitro* Diagnostic Use

Product Code: 8008C

A Rapid Visual Test for the Qualitative Detection of Infectious Mononucleosis Heterophile Antibody.

INTENDED USE

The Chemtrue® Mono Rapid Test is a rapid test for the visual, qualitative detection of heterophile antibodies specific to Infectious Mononucleosis (IM) in human serum, plasma or whole blood. This test kit is intended as an aid in the diagnosis of IM in patients with characteristic clinical symptoms, and is intended for professional laboratory use only. This information can be used by the physician and the patient for disease management.

SUMMARY

IM is an acute, self-limiting disease caused by the Epstein-Barr virus (EBV). Infection with EBV in early life usually is asymptomatic. However, up to 50% of infection occurring in young adulthood and adolescence will develop clinical manifestations associated with IM.
Diagnosis of IM is based on the evaluation of characteristic clinical symptoms and serological changes. Serological diagnosis of IM has been demonstrated by the detection of heterophile and EBV specific antibodies. The heterophile antibody is detectable at some point during IM in most adults. It is a widely accepted practice among physicians to use the detection of heterophile antibodies as an aid in the diagnosis of IM. The Chemtrue® Mono Rapid Test utilizes bovine erythrocyte extract which has a higher sensitivity and specificity than extracts from other species.

PRINCIPLE

The Chemtrue® Mono Rapid Test has been designed to detect IM through visual interpretation of color development in the test device, which is a sandwich solid phase gold conjugate immunoassay. The test device contains a membrane strip which is pre-coated with heterophile antigens on the test region and goat anti-mouse antibody on the control line region. The anti-human IgM antibody-colloidal gold conjugate pad is placed at the end of the membrane. A mixture of colloidal gold conjugate together with the sample and test buffer will move along the membrane chromatographically by capillary action. When the IM heterophile antibodies are present in the patient sample, the mixture will migrate to the test region and form a visible red line as the antibody complexes with the heterophile antigen. When IM heterophile antibodies are absent from the sample, no visible red line will form on test region. Therefore, the presence of a red line on the test region indicates a positive result. A red line will always appear at the control region. This control line serves as a procedural indicator for the proper performance of the test and the device.

REAGENTS AND MATERIALS SUPPLIED

- Individually wrapped test devices with disposable transfer pipette. Each test cassette contains one test strip with heterophile antigen coated membrane and colored antibody pad.
- Test Buffer: 0.1 M Phosphate Buffered Saline, with additives and 0.1% sodium azide.
- Test Instruction.

MATERIALS REQUIRED BUT NOT SUPPLIED

- Serum: Vacutainer tubes
- Whole blood or plasma: EDTA, heparin or citrate
- Finger stick blood: Finger lancet
- Timer

STORAGE AND STABILITY

The test kit is to be stored at room temperature in the sealed pouch for the duration of the shelf-life.

PRECAUTIONS

- *FOR IN-VITRO DIAGNOSTIC USE ONLY.*
- Do not use the test kit beyond the expiration date.
- Do not mix reagents from different lots.
- All patient samples should be treated as if capable of transmitting disease.
- Do not use whole blood stored for more than three days.
- Grossly hemolyzed samples should not be used.
- Test Buffer contains sodium azide. Sodium azide may react with lead or copper plumbing to form potentially explosive metal azides. When disposing of these solutions, always flush with copious amounts of water to prevent azide buildup.

SPECIMEN COLLECTION AND STORAGE

Fingerstick:

- Clean the area to be lanced with an alcohol swab.
- Squeeze the end of the fingertip and pierce with a sterile lancet.
- Wipe away the first drop of blood with sterile gauze or cotton. Allow the second drop to flow directly into the sample well of the test device or use the transfer pipette provided to obtain fresh blood. Add two (2) drops into the sample well.

Whole Blood

- Collect whole blood into a purple, blue or green top collection tube (containing EDTA, citrate or heparin, respectively) by venipuncture.

- The whole blood may be used for testing immediately or may be stored at 4-8℃ up to three days.

Plasma

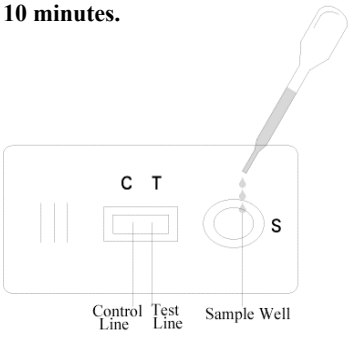
- Collect whole blood into a top collection tube (containing EDTA, citrate or heparin. respectively) by venipuncture.
- Separate the plasma by centrifugation.
- Carefully withdraw the plasma for testing or label and store at 4-8℃ for up to two weeks. Plasma may be frozen at –20℃ for up to one year.

Serum

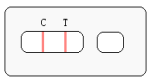
- Collect whole blood into a red top collection tube (containing no anticoagulants) by venipuncture.
- Allow the blood to clot and separate serum by centrifugation.
- Carefully withdraw the serum for testing, or label and store at 4-8℃ for up to two weeks. Serum may be frozen at –20℃ for up to one year.

TEST PROCEDURE

- Review specimen collection instructions.
- Test device, Test Buffer, patient's samples should be brought to room temperature prior to testing. Do not open pouches until ready to perform the assay.
- Remove the test device from its protective pouch. (Bring the device to room temperature before opening to avoid condensation of moisture on the membrane). Label the device with patient identification.
- Add specimen to sample well:
  1) For fingertip blood: Allow the second drop to flow directly into the sample well of the test device or use the pipette provided to obtain fresh blood. Add two (2) drops into sample well
  2) For whole blood samples in collection tubes: add two drops into the sample well, holding the provided transfer pipette in a vertical position.
  3) For plasma or serum samples: add 1 drop of serum or plasma into the sample well, holding the provided transfer pipette in a vertical position.
- Immediately add 2-3 drops of Test Buffer.
- After adding the Test Buffer wait for the red lines to appear. Depending on the concentration of heterophile antibodies present, positive results may be observed within 3 minutes. However, to confirm a negative result, the complete reaction time of 5 minutes is required. **Do not read test results after 10 minutes.**

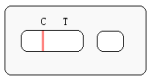


INTERPRETATION OF RESULTS



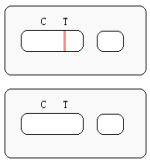
Positive:

Two red lines appear. One red line in the control region **(C)** and one in the test region **(T)**. When testing with strong positive samples, the intensity of the control line may be lighter than expected. Comparison of the line intensities is not recommended.



Negative:

Only one red line appears in the control region. No apparent faint red line on the test line region (T).



Invalid

The test is invalid if the control line is not visible at five minutes. The test failed, or the test procedure was not followed properly. Verify the test procedure and repeat the test with a new testing device.

LIMITATION OF PROCEDURE

- This test kit is to be used for the qualitative detection of IgM antibodies to IM heterophile antigen. A positive result suggests the presence of IgM antibodies to heterophile antigen.
- This test kit should be used for symptomatic individuals suspected of having IM. Diagnosis of IM should be made by confirmation with other clinical findings.
- A negative result does not rule the out possibility of IM because the antibodies to heterophile antigen may be absent or may not be present in sufficient quantity to be detected.

QUALITY CONTROL

Internal Procedural control

- A procedural control is included in the test. A red line appearing in the control region (C) is considered as an internal procedural control, indicating proper performance and reactive reagents.

- A clear background in the result window is necessary. However, when whole blood samples are tested, the background may appear slightly reddish due to a low level hemolysis of some red blood cells. This is acceptable as long as it does not interfere with the reading of the test. The test is invalid if the background fails to clear and obscures the reading of the test result.

EXPECTED VALUES

- During the acute phase of IM, heterophile antibodies are detectable in 80-85% of patients. Heterophile

antibodies are detectable during the first month of illness and decrease rapidly after four weeks.

- Positive results may be persistent for months or even years.
- Some segment of the population who contract IM do not produce measurable levels of heterophile antibodies. Approximately 50% of children under 4 years old who have IM may test negative.

PERFORMANCE CHARACTERISTICS

Sensitivity:

- Since there is no sensitivity standard established for IM heterophile antibodies, the following dilution (test sensitivity) studies were performed for comparison purposes.
- Two mono positive human sera purchased from suppliers were used for the serial dilution in a mono negative human serum.
- The results of the test sensitivity study are summarized in Tables 2a and 2b.
- **Table 2a. Summary of Test Sensitivity / Dilution Study of Serum#1**

Serum#1 Dilution Factor	Mono Test	Mono Test
1:10	+	+
1:20	+	+
1:30	+	+
1:40	+	+
1:50	+/-	+/-
1:60	-	-

- **Table 2b. Summary of Test Sensitivity / Dilution Study of Serum#2**

Serum#2 Dilution Factor	Mono Test	Mono Test
1:10	+	+
1:20	+	+
1:30	+/-	+/-
1:40	-	-

As indicated in Tables 2a and 2b, different positive sera have different titers. Sensitivity of Chemtrue® Mono Rapid Test is very similar to the commercially available test kit as determined by dilution studies.

Accuracy

The accuracy of the Chemtrue® Mono Rapid Test was evaluated in comparison to a commercially available qualitative color immunochromatographic assay for the detection of IM IgM heterophile antibodies in human serum, plasma or whole blood. Four hundred and six human serum, plasma and whole blood samples (288 serum, 103 plasma and 15 whole blood) were used in the comparison study. The comparison results are summarized in Table 1.

Table 1

	Positive	Negative	Total
Commercially available kit Positive	146	2	148
Commercially available kit Negative	3	255	258
Total	149	257	406

The results indicated that the Chemtrue® Mono Rapid Test, demonstrated a Sensitivity of 98.6% (146/148), Specificity of 98.8 % (255/258), and a total agreement of 98.8 % (401/406).

Precision/Site study

The precision of the Chemtrue® Mono Rapid Test has been evaluated at two other sites, including a physician's office and an independent clinical laboratory. Out of 27 positive samples with different levels, all (27) results were positive. Out of 33 negative samples, all (33) results were negative. The results obtained from all site studies demonstrated 100% agreement with the expected result. In addition, one positive and one negative control were tested each day. Results showed 100% agreement for control specimens.

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