***MEDICAL DIRECTOR APPROVAL:*** _____

***SUPERVISOR APPROVAL:*** _____

## SCOPE:

This procedure applies to testing performed in the laboratory section of HHH. The testing procedure, equipment, and data generated are subject to all policies and procedures of the Laboratory Quality Plan and Laboratory General Procedure Manual as well as facility generated Policies and Procedures.

## PURPOSE:

The diagnosis of infectious mononucleosis (IM) is suggested on the basis of the clinical symptoms of fever, sore throat and swollen lymph glands. The highest incidence of symptomatic IM occurs during late adolescence (15-24 years of age). Infectious mononucleosis is caused by the Epstein-Barr Virus (EBV) . The laboratory diagnosis of IM is based on the detection of IM heterophile antibodies. These heterophile antibodies are directed against antigens found in bovine, sheep and horse erythrocytes. The Immuno*Card* STAT! Mono Test utilizes an extract of bovine erythrocytes to give the required sensitivity and specificity.

## PRINCIPLE:

The ImmunoCard STAT Mono Test uses color immunochromatographic dipstick technology with bovine erythrocyte extract coated on the membrane. In the test procedure, serum or plasma is mixed with the diluent. Then the test stick is placed in the mixture and the mixture migrates along the membrane. If the specific Infectious Mononucleosis heterophile antibody is present in the sample, it will form a complex with the bovine erythrocyte extract conjugated color particles. The complex will then be bound by bovine erythrocyte extract immobilized on the membrane and a visible blue test line will appear to indicate a positive result.

## SPECIMEN COLLECTION AND HANDLING:

**Conditions for patient preparation:**

No patient preparation is necessary.

**Specimen type:**

**Venous blood collection:**
Collect venous samples via syringe or collection tube containing EDTA, heparin, SST or red top tube. Follow proper collection for each tube type.
Serum/plasma- centrifuge tube and perform testing on serum either the same day or up to 48 hours after refrigeration at 2 to 8°C.
Serum and plasma specimens held for longer times should be frozen (below -10°C) and tested within 3 months.
Specimen must be at room temperature when testing.

**Specimen labeling:**
All specimens must be properly labeled with patient's first and last name and the date of collection. The collector's initials and the time the blood was drawn are also required for all inpatients.

**Specimen rejection:**
Clots in EDTA sample tube; tubes with incorrect or missing label information; grossly contaminated or leaking tubes; tubes that are QNS for testing (EDTA tubes should be ***filled to the line*** to be processed). For unacceptable specimens, the technologist will notify the physician or their representative of the cancellation and cancel the test in the LIS documenting who was notified. Results that do not correlate with the patient (abnormal results inconsistent with patient condition) should be recollected. The technologist should notify the physician or their representative of the need to recollect the specimen, cancel the test in the LIS, and footnote the notification in the cancellation field.

## EQUIPMENT AND MATERIALS:

**Materials:**

Immunocard STAT!® Mono kit which includes:
Test Sticks
Test Tubes
Transfer pipet
Diluent (contains buffer with 2% sodium azide)
Mono positive control (contains rabbit anti-beef stroma in tris buffer with 2% soium azide and 0.05% gentamycin sulfate preservatives)
Mono negative control (contains goat albumin in tris buffer with 2% sodium azide)
Work station

**Materials not provided in kit:**

Timer

## QUALITY CONTROL:

**Internal Control**
The ImmunoCard STAT! Mono Test provides two levels of internal procedural controls with each test procedure. Results from these internal quality controls must be documented with each sample tested.

- The red Control Line is an internal positive procedural control. The Test Stick must absorb the proper amount of test material and be working properly for the red Control Line to appear.
- A clear background is an internal negative procedural control. If the test has been performed correctly and the Test Stick is working properly, the background will clear to give a discernable result.

If the red Control Line does not appear, the test may be invalid. If the background does not clear and interferes with the test result, the test may be invalid. **Call Meridian Bioscience Technical Services if you experience either of these problems. 800-343-3858. Do not report patient results.**

**External Control**
The external Quality Control material provided in each box will be performed and recorded whenever a new box (kit) is opened and upon training of a new operator.

**Whenever a new kit lot number is put into use, crossover testing using the positive and negative controls from the kit previously in use must be performed. The latex reagent from the new kit must be tested against the positive and negative control from both the previous kit and its provided QC material.**

## PROCEDURE:

### Addition of Specimen:

*For serum, plasma or whole blood samples in tubes:*

Use the Transfer Pipette provided and add one drop to the Test Tube.

- Slowly add 1 drop of Diluent to the bottom of the Test Tube.
- Mix
- Remove the Test Stick(s) from the container. Re-cap the container immediately.
- Place the Absorbent End of the Test Stick into the treated sample. Leave the Test Stick in the Test Tube.
- Read results at 5 minutes. Positive results may be read as soon as the red Control Line appears.

Discard used test tubes and Test Sticks in the suitable biohazardous waste container.

## RESULTS:

**Notes**

A blue or red line, which appears uneven in color density, is considered a valid result.

**Positive**

A blue Test Line and a red Control Line is a positive result for the detection of infectious mononucleosis heterophile antibody. Note that the blue line can be any shade of color and can be lighter or darker than the line in the picture.

**Negative**

A red Control Line but no blue Test Line is a negative result. No infectious mononucleosis heterophile antibody has been detected.

**Invalid**

If after 5 minutes, no red Control Line appears or background color makes reading the red Control Line impossible, the result is invalid. If this occurs, repeat the test on a new Test Stick or call Meridian Bioscience Technical Services at 800-343-3858.

Results from both patient testing and controls must be recorded on the Mononucleosis Testing Quality Control and Patient Testing Log located in the Kits Testing Binder in the Hematology area. Results of crossover testing, when performed, must also be documented.

A result of positive or negative should be entered into the LIS database under workstation 800 under the patient accession number.

## EXPECTED RESULTS:

A heterophile antibody response is observed in approximately 80-90% of adults and children with EBV-caused IM. This percentage drops to approximately 50% for children under four years of age.
While the incidence of IM reflects wide seasonal, ethnic and geographical variation, a large epidemiological study noted that the highest incidence of symptomatic IM occurred during late adolescence (15-24 years of age).

## LIMITATIONS OF PROCEDURE:

- As with all diagnostic assays, the results obtained by this test yield data that must be used as an adjunct to other information available to the physician.
- The Immuno*Card* STAT! Mono Test is a qualitative test for the detection of IM heterophile antibody.
- A negative result may be obtained from patients at the onset of the disease due to heterophile antibody levels below the sensitivity of this test kit. If symptoms persist or intensify, the test should be repeated.
- Some segments of the population with acute IM are heterophile antibody negative.

## REFERENCES:

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ImmunoCard STAT! Mono Test package insert, Meridian Bioscience,Inc., Genzyme Diagnostics, Framingham, MA 01701 Rev. 3854-2, 04/08