**COAG.TRIAGE.4.0 TRIAGE D-DIMER TEST FOR EDTA WHOLE BLOOD AND PLASMA**

**PRINCIPLE**

The Triage® D-Dimer Test is a fluorescence immunoassay to be used with the Triage Meters for the quantitative determination of cross-linked fibrin degradation products containing D-dimer in EDTA whole blood and plasma specimens.

The test procedure involves the addition of several drops of an EDTA whole blood or plasma specimen to the sample port on the Test Device. After addition of the specimen to the sample port, the whole blood cells are separated from the plasma using a filter contained in the Test Device. The specimen reacts with fluorescent antibody conjugates and flows through the Test Device by capillary action. Complexes of each fluorescent antibody conjugate are captured on a discrete zone resulting in a binding assay.

The Test Device is inserted into the Triage® Meter (hereafter referred to as Meter). The results are displayed on the Meter screen and can be printed. All results are stored in the Meter memory to display or print when needed.

**OWNERS**

Supervisor, Community Hospital South

Manager, Regional Hematology

**SPECIMEN COLLECTION**

A. Patient preparation: No special preparation needed.

B. Specimen type:

1. Whole blood or plasma using EDTA as the anticoagulant. No other anticoagulant is acceptable.
2. Minimum sample requirement of 250 uL.
3. Grossly hemolyzed and/or clotted specimens are unacceptable.
4. Gel separators are unacceptable.

C. Specimen stability:

1. Complete testing as soon as possible after collection.
2. If testing is to be delayed, whole blood or plasma may be stored at room temperature or refrigerated at 4oC for up to 24 hours prior to testing. For longer storage, separate plasma and freeze at -20oC or colder in a non-defrosting freezer.
3. Frozen plasma and refrigerated whole blood or plasma specimens must be allowed to reach room temperature before testing. Mix Patient specimens thoroughly:
4. Mix whole blood specimens by gently inverting the tube several times before testing.
5. If possible, mix plasma specimens by vortexing the tube before testing.

**EQUIPMENT**

1. Biosite Diagnostics Triage® D-Dimer Test Kit

Kit contains 25 Test Kit

Test Device 25 each

Transfer Pipettes 25 each

Reagent Code Chip 1 each

Printer Paper 1 roll

1. Triage® MeterPro or Triage® MeterPlus

**REAGENTS**

The Triage® D-Dimer Test Device contains all of the reagents necessary for the quantification of cross-linked fibrin degradation products containing D-dimer in EDTA-anticoagulated blood or plasma specimens.

1. Composition:
2. Murine monoclonal antibodies against D-dimer
3. Fluorescent dye
4. Solid phase
5. Stabilizers
6. Storage and stability:
7. Store the Test Devices in a refrigerator at 2oC – 8oC (35oF – 46oF).
8. Once removed from refrigeration, the pouched Test Device is stable for up to 14 days, but not beyond the expiration date printed on the pouch.
9. Do not remove the Test Device from the pouch until ready for use.
10. Before using refrigerated Test Devices (2oC – 8oC), allow individual foil pouched Test Devices to reach room temperature; this will take a minimum of 15 minutes. If a kit containing multiple Test Devices is being removed from refrigeration, allow the kit box to reach room temperature before use; this will take a minimum of one hour.
11. If the Test Device is not used on the same day of removal from refrigeration, use a soft, felt tip marker to gently write the date of removal from the refrigerator and the date to discard on the foil pouch and/or the kit box.

**CALIBRATION**

When a new lot of Test Devices is opened, the calibration and expiration data for that lot of Test Devices must be transferred to the Meter before patient testing. Use the Reagent Code Chip module supplied with the new lot of Test Devices to transfer the data to the Meter.

1. Frequency: Perform one time for each new lot of Test Devices.
2. Procedure:
3. From the Main Menu, select **<Install New Code Chip>**. Press <**Enter**>.
4. Place the Reagent Code Chip module into the lower left front corner of the Triage Meter and follow the prompts on the screen.
5. Remove the Reagent Code Chip module from the Triage Meter when data transfer is complete.
6. Analyze both levels of external QC after a new Reagent Code Chip has been entered and prior to patient testing.

**Calibration Verification**

Total Calibration Verification is used to verify the calibration of the test devices throughout the measureable range.

Alere recommends running Calibration Verification samples every 6 months.

The order number for the Total Calibration Verification set is 88755. The Total Calibration set contain 2 vials of 5 levels X 0.25mls. The Total Calibration Verification Set is stored at -20oC or colder in a non defrosting freezer. Do not store near the door and do not refreeze material.

**FOR EACH DEVICE TYPE:**

1. Remove 1 set of 5 levels of Calibration Verification samples from freezer.
2. Thaw at room temperature (19oC to 25oC) for 30 minutes. Use within one hour of removal from frozen storage.
3. Mix the control thoroughly by vortexing or inversion prior to testing.
4. Open a device and label device with the vial lot number (can be obtained from the expected values card or the vial).
5. Hold tube with tip facing upward. Make sure all the liquid is at bottom of the vial.
6. Open vial by twisting the tab at the top. Turn the tube over and dispense entire contents into sample port of the test device. Discard the empty tube.
7. Allow the fluid to absorb into the filter.
8. To run in meter: Highlight **Run Test** on the display panel. Press **Enter** to select.
9. Enter your ID number and press **Enter** (if applicable).
10. Use the arrow key to highlight **Patient Sample**. Press **Enter**.
11. Type in the lot number of the calibration verification vial. Enter 1 for the first sample. Press **ENTER** until you see the message ***Measurement in Progress*** on the meter screen.
12. You may apply the second Calibration Verification sample to a test device a few minutes after the first test device is put into the meter.
13. Review the results printed out by the meter to make sure all the test results are within + 2 SD of the mean.
14. Run the rest of the Calibration Verification samples following the same instructions.
15. **METER-TO-METER VERIFICATION:** If multiple meters are used, take the device from the first meter and run it on the second meter and so on. The first reading should take approximately 15 minutes. Subsequent reading should take approximately 2 minutes each. Once inoculated, each device may be read for up to 30 minutes.
16. If the Cal Ver results are > + 2 SD, remove a 2nd vial, thaw 30 minutes and rerun. If it fails again, call Alere technical service. 1-877-308-8287.
17. Enter all data and information, including all meter serial numbers, device, control and cal ver lot numbers, into the form and email to: William.sbai@alere.com.

**QUALITY CONTROL**

1. QC Simulator
2. Function: The QC Simulator mimics a test device and checks the following instrument functions:
3. Optics/calibration
4. Laser stability
5. Carrier alignment
6. Frequency of use: The QC Simulator provided with the Triage® Meter must be tested daily to verify instrument performance.
7. External Quality Controls
8. The QC material to be used is Triage® Total Control 5, levels 1 and 2.

WARNING: The human source material used to produce this product has been tested for hepatitis B antigen, antibodies to hepatitis C, HIV-1 and -2 and found to be non-reactive when using licensed reagents. Inasmuch as no known test method can offer complete assurance that infectious agents are not present, the reagents and patient samples should be handled as though they are capable of transmitting disease.

1. Frequency of use: External Quality Controls are to be analyzed with each new shipment or lot of product, and at least every thirty days. Controls may be run to verify test performance as needed.
2. Storage and stability:
3. Controls are stable until the expiration date on the box when stored at < -20oC in a non-defrosting freezer where possible.
4. Thaw the controls until room temperature and mix well by inversion prior to use.
5. Controls should be used within one hour of thawing.
6. Do not refreeze control material. Each vial should be used once and then discarded.
7. Internal Quality Controls
8. Each Triage® D-Dimer test device contains two internal positive controls that satisfy routine quality control requirements. These controls indicate that sufficient sample was applied to the panel, that the unbound fluorescent label washed sufficiently from the detection zone, and that the panel was inserted and read properly by the Triage® Meter.
9. An unacceptable result from either control causes a warning message on the Triage® Meter indicating that the test should be repeated. Lack of sample results will be indicated by dashes.
10. If a whole blood sample results in an error message, “Warning: Internal QC Out of Range,” spin the sample down and test the EDTA plasma. If the plasma does not result in an error message, the results are reportable.
11. If the error message “Measurement Failure” is displayed, insufficient sample was added to the device. This could be due to:
12. Improper use of the Biosite provided pipette.
13. A clot or fibrin strands present in the sample.

**PROCEDURE**

1. QC Simulator
2. Upon initial receipt of a QC Simulator, the Simulator data must be loaded into the meter by installing the QC Simulator Code Chip.
3. Switch the Triage Meter on.
4. Select **<Install Code Chip>** using the up/down keys from the Main Menu. Press **<Enter>**.
5. Install the selected code chip in the code chip reader port located in the lower left front corner of the Triage® Meter. Press **<Enter>.**
6. The code chip information is automatically installed into the meter memory.
7. Remove the code chip from the port.
8. If installation is unsuccessful, call Biosite Technical Service.
9. Perform Simulator test.
10. At the Main Menu, select **<Run Test>** and **<Enter>.** Select **<QC Simulator>** and press **<Enter>.**
11. At the prompt **<Insert QC Simulator into meter>**, insert the QC Simulator into the meter until it clicks and press **<Enter>.**
12. The device will be drawn into the meter.
13. PASS or FAIL results will appear on the screen and the meter will release the QC Simulator upon test completion; results will automatically print.
14. Remove the QC Simulator from the meter and return it to the black storage box.

NOTE: The QC Simulator is light sensitive and must be stored in the black box provided.

1. Record the Simulator results on the Triage® Meter Daily Maintenance log.
2. If any of the tested functions fail, clean the QC Simulator with canned air or a lint free cloth and repeat the test. If the failure continues, contact Biosite Technical Service. If any of the QC Simulator checks fail, no patient or QC samples can be tested.
3. QC SAMPLE
4. Install the Reagent Code Chip, if necessary.
5. The chip does not need to remain in the port.
6. Install the QC Code Chip.
7. Remove the QC Code Chip from the meter port, and store it with the remaining frozen controls.
8. Log the QC sample onto the appropriate log sheet, if applicable.
9. Open the pouch, remove the test device and label with an identifier.
10. Add well-mixed quality control material into the sample chamber using the transfer pipette provided. To pipette, depress the larger (top) bulb completely and insert the tip into the specimen. Release the bulb slowly. The tube should fill completely with some fluid overflowing into the smaller overflow bulb.
11. Place the tip of the pipette into the sample port of the device and depress the larger bulb completely. The entire amount of sample in the pipette tube must be dispensed. The sample in the smaller overflow bulb will not be expelled.
12. Discard the transfer pipette into the biohazard trash.
13. At the Main Menu, select **<Run Test>** and **press <Enter>.**
14. Select **<QC Sample>** and **press <Enter>.**
15. Enter the QC lot number from the label on the side of the vial, not from the side of the box, using numbers only. To correct if necessary, press **<Delete>** and retype the correct numbers. Press **<Enter>.**

NOTE:

1. External QC samples must be run in pairs. Both Level 1 and Level 2 are to be run consecutively. The meter will not recognize that QC is valid if only one level is run and the NUMBER OF CONTROLS setting is set to 2.

2. Do not re-insert the reagent code chip after QC has been run. Inserting the reagent code chip resets the QC Sample timer and QC samples will need to be repeated.

3. If the NUMBER OF CONTROLS setting is 2 and the 2nd level is rerun, the meter resets the counter and both levels must be run again. Ex. Level 1 passes, Level 2 passes but technician decides to run Level 2 again, the meter will reset the counter with the 2nd run and require Level 1 to be run again.

12. Insert the test device gently into the Triage® Meter until a click is heard.

NOTE: Once sample has been pipetted into a device, the device must be inserted into the meter within 30 minutes. If for any reason there is a delay, the test must be set up again using a new test device.

13. The results will be available in approximately 15 minutes.

14. The test device will be released from the analyzer upon test completion. Discard the test device appropriately. Up to four additional devices may be set up within the first 15 minutes of the initial device’s insertion.

15. Results may be read on the display screen or from the meter printout. Document results appropriately.

16. If the result is backlit on the screen or printout, then the QC sample result is greater than +/-2 SD and must be repeated. If the External QC continues to fail, contact BIOSITE Technical Service.

17. Reagent Lots – QC: A list of Reagent Code Chips in memory, along with corresponding expiration dates for QC samples can be recalled from memory. The list will contain the Lot number, Panel type and expiration date, (if the reagent lot expires prior to the QC sample, the reagent lot expiration date will be displayed).

a. If the QC expiration date has passed, the date will be in reverse video.

b. If the QC for the lot has failed, “FAILED” will appear in reverse video.

c. If a Code Chip has been installed for a new lot, but QC has yet to be established, the words “NOT RUN” will appear in place of this expiration date.

d. To access the information at MAIN MENU, select recall results, QC results, Reagent Lots – QC.

1. PATIENT SAMPLE
2. Log patient information onto the appropriate log sheet, if applicable.
3. Open the pouch, remove the test device and label it with a patient identifier.
4. Add well-mixed EDTA whole blood or plasma into the sample chamber using the transfer pipette provided. To pipette, depress the larger (top) bulb completely and insert the tip into the specimen. Release the bulb slowly. The tube should fill completely with some fluid overflowing into the smaller overflow bulb.
5. Place the tip of the pipette into the smaller port of the device and depress the larger bulb completely. The entire amount of sample in the pipette tube must be dispensed. The sample in the smaller overflow bulb will not be expelled.
6. Discard the transfer pipette into the biohazard trash.
7. At the Main Menu, select **<Run Test>** and **press <Enter>.**
8. Select **<Patient Sample>** and **press <Enter>.** If correct, press **<Enter>.**
9. Confirmation of the correct entry is required. If correct, press **<Enter>.** If incorrect, select correct patient ID, and press (ENTER). Then press **<Delete>** and retype the numbers. Press **<Enter>.**
10. Insert the test device gently into the Triage® Meter until a click is heard.

NOTE: Once sample has been pipetted into a device, the device must be inserted into the meter within 30 minutes. If for any reason there is a delay, the test must be set up again using a new test device.

1. The results will be available in approximately 15 minutes.
2. The test device will be released from the analyzer upon test completion. Discard the test device appropriately. Up to four additional devices may be set up within the first 15 minutes of the initial device’s insertion.
3. Results may be read on the display screen or from the meter printout. Document results appropriately.
4. If the device’s Internal QC results are unacceptable, the patient result will not be displayed. A warning will appear on the display and printout. To troubleshoot, see QUALITY CONTROL, Section C.

**CALCULATIONS**

None

**REPORTING RESULTS**

1. Reference Range: None: see interpretive report
2. Critical Range: Not applicable
3. Sunquest Computer QLS

Function: MEM FUNCTION: 3,3,1

Worksheet: Specific to Site WORKLIST: Specific to Site

Method: Specific to Site

Test Code: DIMRW TEST CODE:

1. Online entry:

Function: OEM

Device: (method/device specific for site)

1. Report patient results in whole numbers.
2. Report D-Dimer results less than 100 ng/mL as <100.
3. Report D-Dimer results of greater than 5000 ng/mL as >5000.
4. An interpretive report will be attached to each result:

<400 ng/ml D-DU

**The D-dimer result can be used as an aid in diagnosis, and should not be used for the exclusion of deep vein thrombosis and/or pulmonary embolism. There have been no established cut-off values for excluding DVT and PE for this method.**

>400 ng/ml D-DU

**The D-dimer result can be used as an aid in diagnosis, and should not be used for the exclusion of deep vein thrombosis and/or pulmonary embolism. There have been no established cut-off values for excluding DVT and PE for this method. A diagnostic radiologic study should be performed in patients**    **suspected to have thromboembolic disease. Increased levels of D-dimer can be associated with a PE, DVT, DIC, malignancies, inflammation, sepsis, surgery, trauma, pregnancy and advancing patient age.**

1. If either the HIS or LIS is unavailable, see the Laboratory Computer Downtime Policy.

**PROCEDURE NOTES**

1. When testing cannot be performed, follow the appropriate back up policy.
2. Notification of instrument downtime should be made to MACL Client Response, the nursing unit and the backup laboratory receiving specimens.

**LIMITATIONS**

1. AMR: 100 – 5,000 ng/mL
2. The results of the Triage® D-Dimer Test should be evaluated in the context of all the clinical and laboratory data available. If Triage® D-Dimer Test results do not agree with the clinical evaluation, additional tests should be performed.
3. This test has been evaluated with whole blood and plasma using EDTA as the anticoagulant. Serum and blood or plasma specimens obtained using other anticoagulants (e.g. heparin or citrate) have not been evaluated and should not be used.
4. There is the possibility that factors such as technical or procedural errors, as well as additional substances in blood specimens that are not listed in the D-Dimer product insert, may interfere with the test and cause erroneous results.
5. Interfering substances: The compounds listed below when added to EDTA anticoagulated plasma did not interfere with the recovery of D-dimer

Compound Concentration Tested

Hemoglobin 500 mg/dL

Lipids 3000 mg/dL

Bilirubin 15 mg/dL

Fragment D 20 µg/mL

Fragment E 20 µg/mL

1. Severely hemolyzed specimens should be avoided.
2. The hematocrit was varied between 30% and 55% with no significant effect on the recovery of D-dimer. RA factor has not been tested.

**REFERENCES**

1. Triage® D-Dimer Product Insert; 22677 Rev E, Biosite Incorporated, 2009.
2. Triage® MeterPro Users Guide, 22719 Rev C, 2007/02/09.
3. Triage® Total Controls 5 Product Insert, Rev E, 2009.