|  |  |
| --- | --- |
| PurposePrinciple | This procedure provides instructions forperforming the D-Dimer assay.The specific degradation of fibrin (i.e., fibrinolysis) is the reactive mechanism responding to the formation of fibrin. Plasmin is the fibrinolytic enzyme derived from inactive plasminogen. Plasminogen is converted into plasmin by plasminogen activators. The main plasminogen activators are tissue plasminogen activator (tPA) and pro-urokinase which is activated into urokinase (UK) by, among others, the contact system of coagulation. In the bloodstream, plasmin is rapidly and specifically neutralized by alpha 2 – antiplasmin, thereby restricting its fibrinogenolytic activity and localizes the fibrinolysis on the fibrin clot. On the fibrin clot plasmin degrades fibrin into various products, (i.e., D-Dimers). Antibodies specific for these products, which do not recognize fibrinogen, have been developed. The presence of these various fibrin degradation products, among which D-Dimer is the terminal product, is the proof that the fibrinolytic system is in action in response to coagulation activation.When a beam of monochromatic light is allowed to transverse a suspension of microlatex particles to which specific antibodies have been attached by covalent bonding and if the wavelength of the light is much greater than the diameter of the latex particles, the light is only slightly absorbed. In the presence of the antigen being tested for, the antibody-coated latex particles agglutinate to form aggregates of a diameter greater than the wavelength of the light; more of the latter is absorbed. This increase in light absorption is a function of the antigen level present in the test sample. |

|  |  |
| --- | --- |
| Scope | This procedure is intended for Clinical Laboratory Scientist (CLS) and Medical Laboratory Technicians (MLT) who are trained and competent in performing the D-Dimer assay.This Procedure replaces LCoM 219 STA-R Evolution Quality Control and LCoM 221 STA-R Evolution Sample Processing. |

|  |  |
| --- | --- |
| Specimen sources | Whole Blood collected in 9:1 ratio of blood in a 3.2% (0.109M) Sodium Citrate tube |

|  |  |
| --- | --- |
| Specimen collection | Refer to LPhM 200 Performing Venipuncture at KP LAMC |

|  |  |
| --- | --- |
| Specimen transport | Refer to LSPM 203 Specimen Collection, Handling, Packaging and Transportation |

|  |  |
| --- | --- |
| Specimen storage | Whole blood or plasma can be stored at 20 + 5◦ C for 8 hours.Do not store whole blood or plasma at 2-8◦ C |

|  |  |
| --- | --- |
| Specimen rejection | Reject specimen under any of the following conditions:* Clotted
* Samples collected in wrong collection tube
* Short draws and over draws
* Samples with visible hemolysis
* Samples with unusually shortened results must be checked for clot
 |

|  |  |
| --- | --- |
| Equipment | * Stago STA-R Evolution
* Centrifuge

\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ |
|  |  |
| Material and**Supplies** | * Pipettes
* Pipette tips
* Cuvette Roll-1000
* Magnetic Stir Bars
* STA® Reducer
* Reagent Grade Water
* STA – DESORB U
* STA® - Cleaner Solution

\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ |

|  |  |  |  |
| --- | --- | --- | --- |
| Reagent/s | **Description** | **Preparation** | **Stability** |
| STA® Liatest® D-Dimer:**Reagent 1:** Tris Buffer**Reagent 2:** Latex | Ready to use. Allow the reagent to stand at room temperature for 15 minutes. Mix gently without creating bubbles. Then, place a STA**®** mini Reducer and the perforated cap on the vial perforated plastic cap on the vial.Write the date and initial on the vial before loading on the instrument.Ready to use. Allow reagent to stand at room temperature for 15 minutes. Mix gently without creating bubbles. Then, place a STA**®** mini Reducer and the perforated cap on the vial perforated plastic cap on the vial. Write the date and initial on the vial before loading on the instrument. | 15 days on the STA-R Evolution®15 days on the STA-R Evolution® |
|  | STA® - Owren-Koller Buffer | Ready to use buffer. Used by the STA-R Evolution® to perform dilutions of controls and patients’ plasmas.Write the date and initial on the vial before loading on the instrument. | 72 hours on the STA-R Evolution® |

|  |  |
| --- | --- |
| Safety | All laboratory employees are expected to maintain a safe working environment and an injury-free workplace. Laboratory employees are responsible for their own safety, and the safety of others and adhering to all departmental and medical center safety policies and procedures.* For standard precautions and safety practices in the laboratory; see LGM

8000, specifically, but not limited to, equipment safety, proper body  mechanics, sharps exposure and proper use of personal protective  equipment (PPE).* For Universal Body Substance precautions, see LGM 8005, specifically, but not limited to, exposure to body fluids.
* For proper handwashing, see LGM 8010, specifically, not limited to, proper handwqashing.
* For proper infection control, see LGM 8004, specifically, but not limited to, proper use of gloves.
* For proper handling of regular and infectious waste, see LGM 8006, specifically, but not limited to, proper disposal of regular and biohazardous waste.
* For proper cleaning of work area, see LGM 8007 – Cleaning Work Areas.
* For proper handling of chemicals and reagents, see the Chemical Hygiene Plan.
* For proper storage and disposal of chemical hazardous waste, see LGM 8012.
 |
|   |  |

**Calibration/** The kit reagents are pre-calibrated: this calibration is identical for all the reagents

**Verification** of each lot.

 Entering the data for the calibration curve: The database of the STA-R

 Evolution® monitors all reagent lot numbers. When the operator scans a new lot

 of Liatest D-Dimer reagent, STA-R Evolution® will request the operator to scan

 the bar code printed on the bar code insert across the STA–R®/STA-R Evolution®

 bar code reader.

* Follow the steps below to view the calibration curve.

|  |  |
| --- | --- |
| **Step** | **Action** |
| 1 | Click on the Calibration menu icon. |
| 2 | Click **DDIM TEST** (on the test abbreviation) |
| 3 | Calibration screen includes current calibration curves and product information |
| 4 | Positions for two different lots of reagent are available.  |
| 5 | Click on the calibration icon  or calibration icon  |
| 6 | Curve will appear on the STA-R Evolution® screen. Click on the Print icon |

 The calibration curve will be validated for the lot being used once the two Liatest

 D-Dimer controls have been run. If the validation controls are outside the

 assayed range, The STA–R®/STA-R Evolution® will not run patient samples.

 Refer to LCoM 416 STA-R Evolution® Calibration/Verification

|  |
| --- |
| Quality Control |

D-Dimer Controls must be ran at least once per shift using theSTA® Liatest® Control. When Quality Control tolerance limits are exceeded based on the QC

 criteria defined in LGM 2022 Qulaity Control (QC) Policy, corrective action

 must be taken and documented before analyzing

 patient samples.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
|  |

|  |  |  |
| --- | --- | --- |
| **Controls** | **Preparation** | **Stability** |
| STA® Liatest® Control N+P | Reconstitute each vial with 1.0 ml reagent grade water. Let sit 30 minutes at room temperature. Swirl gently. Write the date and initial on the vial after reconstitution. | Reconstituted stability on the STA-R Evolution® is 8 hours. |

 |
|  | * Follow the steps below to run D-Dimer Quality Control
 |
| Step | Action |
| 1 | Click the icon |
| 2 | Click the **DDIM** test abbreviation |
| 3 | Click the **Control Level #** tab (#corresponds to the number of the level) |
| 4 | Click the button |
| 5 | Confirm by clicking **OK**  |
| 6 | Repeat for each level |
| 7 | All control ranges are monitored automatically by the STA–R Evolution®. Control results are automatically filed in the STA–R Evolution® QC file. All results for a 24-hour period will be converted to a “mean” value at midnight. This mean is used in the statistical data and is plotted on the Levy-Jennings chart as a daily mean. Each point can be viewed on the Levy-Jennings Daily control chart by clicking on the left arrow.  |

* Follow the step below to Display Daily Controls

|  |  |
| --- | --- |
| Step | Action |
| 1. | Click on the The **Daily controls** screen is displayed:   |
|  | * For Quality control result >2SD, the value will be flagged ‘To Validate’. ‘To Validate’ denotes that the result must be reviewed and accepted or appropriate action taken such as repeating the quality control run. Follow the step below to validate the quality control result.
 |
| Step | Action |
| 1 | From the **Daily Control** **screen**, **double click** on the quality control to be validated. **The following options will display:*** **Validate**
* **Rerun**
* **Delete**
 |
| 2 |

|  |  |
| --- | --- |
| **If…** | **Then…** |
| The quality control result is acceptable | Choose ‘Validate’ |
| The quality control requires repeat or rerun | Choose ‘Rerun’ |
| The quality control needs to be excluded from the daily statistical data. | Choose ‘Delete’ |

 |

* + - * Follow the step below to print daily Quality Control results.

|  |  |
| --- | --- |
| Step | Action |
| 1. | From the **Daily Control screen**, **click** on  to print daily quality control results. |

|  |  |
| --- | --- |
| Procedure | * Follow the steps below to run D-Dimer on patient samples.
 |
| Step | Action |
| 1 | Place tubes in rack |
| 2 | Place rack(s) on tray |
| 3 | Place tray on STA-R. Tubes are automatically loaded, their barcoded labels are read, work lists are downloaded from the host computer or the predefined profile is applied according to the chosen option. Tests are automatically carried out by the analyzer. |
| 5 | **Test Panel** Click on  |
| 6 | If the instrument is unable to read the barcode, the automatic unloading of not read tubes is selected  and all tubes which are not read are automatically unloaded.The **Manual Input of Patient ID’s** window is displayed for re-identification. |
| 7 | Enter the sample ID |
| 8 | Click on to load the rack |

**Results**

**Reporting** **For D-Dimer DVT and D-Dimer PE:**

* + - * If the result for D-Dimer DVT or D-Dimer PE is >Vmax, report the test as >4000. Do NOT dilute.
			* If the result for D-Dimer DVT or D-Dimer PE is below 270, the result should be reported as <270

 **For D-Dimer DIC:**

* + - * If the result for D-Dimer DIC is >Vmax, program the sample to be diluted 1:5
			* If the 1:5 dilution is not >Vmax, report the value
			* If the 1:5 dilution result is still >Vmax, report >20000
			* If the result for D-Dimer DVT or D-Dimer PE is below 270, the result should be reported as <270

|  |  |
| --- | --- |
| Clinical Significance | Clinical applications for this test are as follows: Disseminated Intravascular Coagulation (DIC), negative predictor for the diagnosis of a thrombotic episode (i.e., DVT, PE), efficacy of treatment for a thrombotic episode and screen for possible re-occurrence (MI), and screen for other activation states of coagulation (i.e., post-operative, cancer, cirrhosis). |
| Reference Range |

|  |  |
| --- | --- |
| D-Dimer DVT Reference Range | <=499 ng FEU/ml |
| D-Dimer PE Reference Range | Age dependent |
| D-Dimer DIC Reference Range | <=499 ng FEU/ml |
| D-Dimer DVT Clinical Reportable Range | 270-4,000 ng FEU/ml |
| D-Dimer PE Clinical Reportable Range | 270-4,000 ng FEU/ml |
| D-Dimer DIC Clinical Reportable Range | 270-20,000 ng FEU/ml |

 |

|  |  |  |  |
| --- | --- | --- | --- |
| Interpretation Results / Critical Values |

|  |  |
| --- | --- |
| D-Dimer Critical Values | None |

 |

|  |  |
| --- | --- |
| Limitations | * Cloudy plasmas may lead to an under-estimation of the D-Dimer level. Ensure that the absorbance value at 540 nm of the plasma diluted 1:6 with STA**®** - Owren-Koller buffer is < 0.35.
* An over-estimation of D-Dimer level may be seen in the following conditions; FDP concentrations greater than 15 *u*g/ml, the presence of rheumatoid factor at a level greater that 50 IU/ml, and the presence of anti-bovine albumin and/or anti-mouse antibodies in certain subjects
* The STA**®** Liatest**®** D-Dimer is insensitive to the following substances: hemoglobin (up to 2 g/l); conjugated bilirubin (up to 290 mg/l); unconjugated bilirubin (up to 200 mg/l); unfractionated heparin (up to 2 IU/ml0), and LMWH (up to 2 anti-Xa IU/ml).
 |

|  |  |
| --- | --- |
| Controlled Documents | The following non-controlled documents support this procedure. |
| Document No. | Name of Documents |
| LPhM 200 | Performing Venipuncture at KP LAMC |
| LSPM 203 | Specimen Collection, Handling, Packaging and Transportation |
| LGM 2022 | Quality Control (QC) Policy |
| LCoM 416 | STA-R Evolution® Calibration/Verification |
| LGM 8000 | Safety Practices |
| LGM 8004 | Infection Control |
| LGM 8005 | Universal Body Substance Precautions |
| LGM 8006 | Handling of Regular and Infectious Waste |
| LGM 8007 | Cleaning Work Areas |
| LGM 8010 | Hand washing Policy |
| LGM 8012 | Storage and disposal of Chemical Hazardous Waste |

|  |  |
| --- | --- |
| Non-Controlled Documents | The following controlled documents support this procedure. |

|  |  |
| --- | --- |
| Document No. | Name of Documents |
|  | STA-*R* Evolution Reference Manual |
|  | STA® Package Inserts |

|  |  |
| --- | --- |
| Author(s) | Rosalie I. Fajardo, MS CLS(ASCP) |