Sutter Health
Sutter Roseville Medical Center

Origination:	*N/A*
Effective:	*N/A*
Final Approved:	*N/A*
Last Revised:	*N/A*
Next Review:	*N/A*
Owner:	*Alex Alba: Spvr, Laboratory*
Policy Area:	*Lab - Coag*
References:	
Applicability:	*Sutter Roseville Medical Center*

Performing VerifyNow PRUTest and Aspirin Platelet Reactivity Testing

Purpose

This document provides instructions on how to use the VerifyNow PRUTest and Aspirin Platelet reactivity test devices to measure platelet induced aggregation.

Policy

- This procedure is to be followed when the VerifyNow test is requested
- In the event that the primary VerifyNow instrument is out of service, samples will be performed on the second VerifyNow instrument as a backup plan for testing
- Prepared VerifyNow blood collection kits are used when the VerifyNow test is requested
- Order code is CLOPB

Principle

- The VerifyNow PRUTest measures the level of platelet P2Y12 receptor blockade. Substances known to block the P2Y12 receptor include thienopyridine drugs, including clodpidogrel (Plavix). The VerifyNow PRUTest is used to monitor the effect of Plavix therapy on platelet function.The VerifyNow Aspirin test measures platelet function based on the ability of activated platelets to bind fibrinogen. Aspirin affects platelet function by irreversibly inhibiting the COX-1 enzyme which ultimately affects activation of GPIIb/IIIA receptors involved in platelet aggregation.The VerifyNow System is a turbidimetric optical detection system. Light transmittance increases as activated platelets bind and aggregate fibrinogen coated beads. The instrument measures changes in optical signal and reports results in P2Y12 Reaction Units (PRU) or Aspirin Reaction Units (ARU), depending on the test cartridge used for testing.

Supplies

- VerifyNow System Analyzer
- VerifyNow PRUTest test device Ref. 85225
- VerifyNow Aspirin test device Ref. 85053
- VerifyNow Assay Wet QC
- 21 gauge needle and hub
- Clear top no additive blood collection tube as discard tube
- Greiner Bio-One Vacuette Partial-Fill 2 ml blood collection tube (3.2% sodium citrate)

- EDTA blood collection tube

Reagents and Storage

- VerifyNow PRUTest and Aspirin Test devices
  - Use prior to expiration date printed on each package
  - Store test devices at 2-25°C
  - If refrigerated, the test device should come to room temp 18-25°C before use
  - Store test devices in foil pouch until ready for use
- VerifyNow Wet Quality Control Level 1 and Level 2
  - Use prior to expiration date printed on each package
  - Store QC material at 15-25°C
  - Control material should be used within 15 min of reconstitution

Specimen Requirements

- 2 Greiner Bio-one partial fill blood collection tubes (3.2% sodium citrate tube)
- 1 EDTA blood collection tube (collect after Greiner tubes)
- Refer to VerifyNow specimen collection and transport SOP for detailed information

Specimen Stability

- PRUTest - Specimen must equilibrate at room temp for a minimum of 10 min from collection time and test must be performed no longer than 4 hrs from collection time
- Aspirin PLT Reactivity test- Specimen must equilibrate at room temp for a minimum of 30 min from collection time and test must be performed no longer than 4 hrs from collection time

Specimen Rejection

- Use of collection tubes other than the blood collection tubes described in the specimen requirements section
- Venipuncture using a needle smaller than 21 gauge
- No additive discard tube not used
- Improperly filled collection tubes
- Clotted or hemolyzed specimens
- Sample with PLT Clumps
- Specimens greater than 4 hours old since collection
- Specimens that were transported in the pneumatic tube system
- Specimens that have been centrifuged, refrigerated, or frozen.

Calibration and QC

- No calibrations are performed by the user on the VerifyNow system
- Electronic QC is run daily and every 8 hrs of patient testing
- Wet QC is run monthly and with every new lot/new shipment of reagents
- Refer to the procedure VerifyNow System: Maintenance and Quality Control for detailed information

Procedure A

Follow the steps below for patient specimen collection

<table>
<tr><th>Step</th><th>Action</th></tr>
<tr><td>1.</td><td>Determine how the blood specimen will be collected:
<table>
<tr><th>If</th><th>Then</th></tr>
<tr><td>Venipuncture</td><td>• Venipuncture is preferred collection method
• Use 21 gauge needle
• Do not use smaller needle than 21 gauge
• Specimen must be transferred immediately to blood collection tubes if straight needle and syringe is used
• 21 gauge butterfly needle should not be used routinely and only used for difficult venipunc</td></tr>
<tr><td>Indwelling catheter</td><td>• Cather must be free from clots
• Specimen cannot be collected from an IV start</td></tr>
</table></td></tr>
<tr><td>2.</td><td>Use the clear to no additive blood collection tube(s) for the discard tube
<table>
<tr><th>If</th><th>Then</th></tr>
<tr><td>Venipuncture</td><td>Discard at least 2 ml</td></tr>
<tr><td>Indwelling Cather</td><td>Discard at least 5 ml</td></tr>
</table></td></tr>
<tr><td>3.</td><td>Collect 2 Greiner blood collection tubes. Fill to the mark on the tube.</td></tr>
<tr><td>4.</td><td>Collect EDTA tube after Greiner tubes.</td></tr>
<tr><td>5.</td><td>Gently invert at least 5 times to completely mix the contents of the tubes.</td></tr>
<tr><td>6.</td><td>Label tubes according to established procedure.</td></tr>
<tr><td>7.</td><td>• Do not transport specimens in pneumatic tube system.
• Deliver samples to laboratory by hand.</td></tr>
<tr><td>8.</td><td>Do not centrifuge, refrigerate, or freeze specimens.</td></tr>
<tr><td>9.</td><td>Receive specimens in the LIS and deliver directly to the VerifyNow testing area.</td></tr>
</table>

Procedure B

Follow the steps below for patient testing

<table>
<tr><th>Step</th><th>Action</th></tr>
<tr><td>1.</td><td>Run the EDTA specimen on a hematology analyzer to obtain HCT and PLT results. Review HCT and PLT results and resolve any instrument flagging for these parameters according to hematology analyzer procedure
<table>
<tr><th>If</th><th>Then</th></tr>
<tr><td>Specimen clotted</td><td>• Do not run Verify Now test devices
• Recollect a specimen</td></tr>
<tr><td>PLT Clumps</td><td>• Verify on slide review and do not run Verify Now test devices
• Recollect a specimen</td></tr>
</table></td></tr>
</table>

	PLT count and HCT results outside of PRU testing limitations • PLT <119 K/uL or >502 K/uL • HCT < 33% or >52%	• Do not run the test • Report as Invalid • Refer to Procedure 2 of the Reporting Results section for detailed instructions
	PLT count and HCT results outside of ARU testing limitations • PLT <92 K/uL • HCT <29% or >56%	• Do not run the test • Report as Invalid • Refer to Procedure 2 of the Reporting Results section for detailed instructions
	Specimen is acceptable	• Proceed to step 2
2.	Let sample equilibrate at room temp for the appropriate minimum amount of time after collection prior to testing, but no longer than 4 hours. • PRUTest: 10 minutes – 4 hours • ARU (Aspirin): 30 minutes- 4 hours	
3.	If the instrument display is not currently on, press any key to turn it on. The instrument should be on for at least 15 minutes prior to use.	
4.	From the Main menu, enter your operator ID and password. Note: Scan your ID badge barcode for your operator ID and password is 123	
5.	From the main screen, press the key next to the patient test icon.	
6.	Using the barcode scanner, scan the patient's LIS barcode. The CID will be the patient ID.	
7.	Open the testing device pouch and remove the test device just before use. Handle test device only by the finger grip.	
8.	When prompted by the instrument: • Remove the needle sheath by pulling directly up on the sheath. Do not twist the sheath as this may remove the needle • Open the sample compartment cover • Insert the device into the instrument device. Press down until it clicks into place.	
	If	**Then**
	The test device is the first from a new lot number of test cartridges	• When barcode screen is displayed, using the barcode scanner, scan the barcode on the bottom of the test device until an audible beep is heard which indicates that the information was read • Proceed to step 9 when test device pouch barcode is read
	If test device is from the current lot of test cartridges	• Proceed to step 9
9.	When prompted with the image of an inverted tube, gently mix the sample by inversion at least 5 times.	
10.	When prompted with the Insert tube icon, insert the sample into the sample well of the test	

	device with the rubber stopper facing downward so that the needle pierces the stopper. The instrument will produce 2 audible beeps when tube has been fully inserted.
11.	Close the sample compartment cover. • The instrument automatically draws the sample from the specimen tube into the test device and will proceed with the analysis
12.	A countdown will be displayed in about 30 seconds and a calculator will be displayed when the test is near completion. DO NOT remove the sample while testing is in progress • PRUTest will display as Verify Now P2Y12 and take about 3 minutes to complete • ARU test takes about 5 minutes to complete
13.	When the result appears on the screen, record the result on the patient testing log. Press key next to printer icon to print result if printer is available.
14.	When prompted, open the sample compartment cover and remove the testing device and sample tube together in one piece by grasping the test device finger grip and pulling straight up then close the sample compartment cover.
15.	Discard the sample and testing device together in a Sharps container. Do not remove the tube from the testing device
16.	Repeat steps 2-15 for additional test devices

Reprinting patient results

Follow the steps below to reprint patient results:

Step	Action
1.	From the main screen, press the key next to the file icon.
2.	Type in the patient CID followed by the Enter (;) key
3.	All results will be displayed.
4.	Press the key next to printer icon to reprint result if printer is available.

Error Codes

The following codes are the most common that are specifically associated with the VerifyNow PRUTest. Consult the Troubleshooting section of the VerifyNow System User Manual for a complete list of error codes and suggested resolutions.

<table>
<tr><th>If</th><th>Then</th></tr>
<tr><td>Error code 24</td><td>Run the EQC on the VerifyNow instrument
<table>
<tr><th>If</th><th>Then</th></tr>
<tr><td>EQC passes. This verifies that the test system is operating normally</td><td>Investigate possible problems with the blood sample
• Improper blood collection technique and/or specimen handling
• Improper or expired tube type
• Discard tube was used to run the test
• Patient has low PLT, low HCT, or an inherited platelet</td></tr>
</table></td></tr>
</table>

		disorder • Patient is on an interfering medication like ReoPro, Integrilin, or Aggrastat • A WQC tube was run in Test mode rather than QC mode • Greiner sample tube is expired
	EQC passes and no problem with the patient sample is found	Run WQC to confirm integrity of the test device and reagents
	EQC does not pass	Consult the Troubleshooting section of the VerifyNow System User Manual for addressing failed EQC. Repeat patient testing when problem is resolved
Error code 28	Run the EQC on the VerifyNow instrument	
	If	**Then**
	EQC passes. This verifies that the test system is operating normally	Investigate possible problems with the blood sample • Improperly mixed blood sample was used for testing • Patient has HCT outside of applicable range (33-52% for PRUTest and 29-56% for Aspirin) • Sample was not run within specified period of time
	EQC passes and no problem with the patient sample is found	Run WQC to confirm integrity of the test device and reagents
	EQC does not pass	Consult the Troubleshooting section of the VerifyNow System User Manual for addressing failed EQC. Repeat patient testing when problem is resolved

Reportable Range

Any value that is outside the reportable range will display as an error and no numerical value will be printed or reported

Parameter	Reportable Range
ARU	350-999
PRU	0-999

Reporting Results

- The order code is CLOPB and includes tests CLOP(PRU), PFASA (ARU), PLT, and HCT
- Patient results are documented on the VerifyNow system patient result log
- PRU and ARU test results are reported as "INVALID" when the following situations occur
  - When the PLT and HCT are outside test performance limits (refer to procedure and test limitation section)
  - When error codes 24 or 28 are generated
- The ordering physician is notified when the PRU test is reported as INVALID

- Patient results are entered manually in the LIS
  - Use worksheet RVKM to report PLT and HCT results
  - Use worksheet RVKM to report CLOP and PFASA results
- Use procedure 1 to report valid VerifyNow results
- Use procedure 2 to report invalid VerifyNow results

Procedure 1 - Follow the steps below to report valid VerifyNow results

Step	Action
1.	In the CLOP field, enter the PRU test result.
2.	In the PFASA field, enter the ARU test result.
3.	Result PLT and HCT manually using RVKM worksheet.
4.	Verify LIS entry and document on patient result log sheet

Procedure 2 - Follow the steps below to report Invalid VerifyNow results

<table>
<tr><th>Step</th><th>Action</th></tr>
<tr><td>1.</td><td>Result CLOP and/or PFASA field with the ETC "INVALD" (Invalid).</td></tr>
<tr><td>2.</td><td>Result PLT and HCT manually using RVKM worksheet.</td></tr>
<tr><td></td><td>
<table>
<tr><th>If</th><th>Then</th></tr>
<tr><td>PLT and/or HCT is a critical value</td><td>• Refer to the Critical Value Reporting procedure to call critical lab result to the provider
• Proceed to step 3</td></tr>
<tr><td>PLT and/or HCT is not a critical value</td><td>• Proceed to step 3</td></tr>
</table>
</td></tr>
<tr><td>3.</td><td>Verify LIS entry and document on patient result logsheet.</td></tr>
</table>

PRUTest results - Two ETC will append automatically to all CLOP results:

- CLOP1 - “Accumetrics has changed the Plavix (P2Y12) test instrument to report only a single PRU value. % inhibition results are no longer reported. Accumetrics states the patients taking Plavix may have results between 18 and 435. This statement is not clinically helpful since that range covers almost the entire reportable range for the test.”
- CLOP12 - “Testing performance may be affected when platelet count is below 119,000 or above 502,000 or when hematocrit is below 33% or above 52%.”

ARU test results - The comments below will auto append to all PFASA results:

- PFASA1- “Testing performance may be affected when Platelet count is below 92000 per microliter or when Hematocrit is below 29 or above 56 percent” will apply to all PFASA results.
- In addition, one of two ETCs will append automatically to all PFASA results dependent on the result:
  - PFASA <550: PFAAIP= Less than 550 ARU, platelet dysfunction consistent with ASA is present, patient is aspirin sensitive
  - PFASA>550:PFAAIN= Greater than 549 ARU, no ASA associated dysfunction has been detected, patient is aspirin resistant

Reference Range

- CLOP=194-418 PRU
- PFASA=620-672 ARU

Critical Limits

- HCT and PLT (refer to Appendix A: Lab Critical Value list)

Procedure and Test Limitations

- VerifyNow testing is not intended for use on patients with inherited platelet disorders. Patients with von Willebrand Factor Deficiency, Glanzmann Thrombasthenia and Bernard-Soulier Syndrome have not been studied
- Results should be interpreted in conjunction with other clinical and laboratory data available to the clinician
- If the PRUTest result is reported as “Interference” (error code 24), valid test results cannot be obtained often due to the presence of an interfering substance such as GP IIb/IIIa inhibitors (abciximab, eptifibatide and tirofiban), other antiplatelet agents (cilastazol), or an inherent platelet defect
- Other classes of commonly used drugs were tested by the manufacturer and found to have no significant effect on the VerifyNow PRUTest. For specific information, consult the manufacturer’s package insert
- According to the product insert, PRUTest performance may be affected by HCT <33 % and >52% or PLT <119,000 and >502,000/uL,
- PRUTest performance is not affected by triglyceride concentrations between 41-824 mg/dL, cholesterol concentration between 98-316 mg/dL, and fibrinogen concentrations 171-599 mg/dL
- VerifyNow ARU (Aspirin) test results may be affected by the presence of other anti-platelet medication besides aspirin. P2Y12 inhibitors (Plavix, Ticlid, Effient), NSAIDs (ibuprofen, naproxen, diclofenac, indocin, feldene), GPIIb/IIIa inhibitors (ReoPro, Integrilin, Aggrastat), and other anti-platelet medications may inhibit platelet function. Patients taking these medications should not be tested on the VerifyNow Aspirin test. For specific information, consult the manufacturer’s package insert
- According to the product insert, ARU (Aspirin) test performance may be affected by HCT <29% and >56% or PLT <92,000/uL
- ARU test performance is not affected by triglyceride concentrations up to 577 mg/dL, and fibrinogen concentrations 164-529 mg/dL
- The performance of the VerifyNow ARU (Aspirin) test with non-drug induced platelet abnormalities is not known

Supporting documents

- Form A: VerifyNow System Patient Test Result Log

Related Procedures

- VerifyNow Quality Control and Maintenance Procedure
- VerifyNow Specimen Collection and Transport Procedure
- Critical Value Reporting Appendix A: Critical Value List

References

- VerifyNow System Operating Manual
- VerifyNow PRUTest Package Insert
- VerifyNow Aspirin Package Insert
- VerifyNow Assay WQC

All revision dates:

Attachments

Form A: VerifyNow system patient result log