Beaumont

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Document Contact:	*Tamara Sabih: Medical Technologist Lead*
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Platelet Function Analysis- DADE PFA-100- RO

Document Type: Procedure

I. PURPOSE AND OBJECTIVE:

A. The DADE PFA-100 platelet function analyzer uses whole blood flowing at a high rate of shear through an aperture in a membrane coated with a combination of collagen and epinephrine or collagen and ADP. This analyzer measures high shear Von Willebrand Factor (VWF) dependent platelet adhesion and aggregation.

B. Blood flows through a defined aperture in a membrane coated with collagen and ADP (cartridge 1), or collagen and epinephrine (cartridge 2). Platelets stimulated by the collagen and agonist accumulate and plug the aperture causing blood flow to cease. The end point of the assay is closure time. Inhibited or diseased platelets will have an abnormally prolonged closure time.

C. Drugs such as aspirin will prolong closure time of the collagen and epinephrine cartridge but not the collagen plus ADP cartridge. Patients with Bernard Soulier Syndrome (BSS), Glanzmann's Thrombasthenia and Von Willebrand's Disease (VWD) will show prolonged closure times with both cartridges.

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II. ACRONYMS:

A. Adenosine-5'-diphosphate (ADP)

B. Closure Time (CT)

C. Collagen (COL)

D. Coefficient Of Variation (CV)

E. Epinephrine (EPI)

F. Platelet Function Analysis (PFA)

III. SPECIMEN COLLECTION AND HANDLING:

Type	Two tubes of Whole blood collected in a 5 mL Hemogard vacutainer (2.7 mL draw).
Anticoagulant	3.2% sodium citrate
Amount	9:1 ratio (2.7 mL whole blood + 0.3 citrate) must be maintained. Tubes must be

	full.
Specimen Handling	**Specimens must NOT be sent through the pneumatic tube system!!! Do NOT spin sample! Whole blood is used for this analysis!**
Timing	Specimens must be kept at room temperature (20-25^{o}C) and analyzed within 4h of collection
Criteria for Unacceptable Specimens	Specimens sent through the pneumatic tube system, containing clots, gross hemolysis, or inappropriate volume are unacceptable and must be redrawn

IV. SUPPLIES/EQUIPMENT

A. DADE PFA-100 Analyzer

B. PFA Vacuum Cartridge

C. PFA Priming Cartridge

D. PFA O-Ring Cleaning Pad

V. REAGENTS:

A. **DADE PFA Collagen/Epinephrine (COL/EPI) Test Cartridge**: A test cartridge unit containing a membrane coated with 2 mcg of equine Type I collagen and 10mcg epinephrine bitartrate. Unopened pouches are stable at 2-8^{o}C until expiration date printed on the label. Test cartridges are stable up to 30 days after opening the pouch when stored at 2-8^{o}C. Test cartridges stored at room temperature (16-26^{o}C) in a sealed or unsealed pouch are stable for up to 4h.

B. **DADE PFA Collagen/ADP (COL/ADP) Test Cartridge**: A test cartridge unit containing a membrane coated with 2 mcg of equine Type I collagen and 50 mcg adenosine-5'-diphosphate (ADP). Unopened pouches are stable at 2-8^{o}C until expiration date printed on the label. Test cartridges are stable up to 30 days after opening the pouch when stored at 2-8^{o}C. Test cartridges stored at room temperature (16-26^{o}C) in a sealed or unsealed pouch are stable for up to 4h.

C. **DADE PFA Trigger Solution:** A trigger solution vial containing 11 mL isotonic saline (0.9% aqueous sodium chloride). Trigger Solution in unopened vial is stable at room temperature (16-26^{o}C) until the expiration date printed on the label. Trigger Solution is stable up to 60 days after the vial is placed on the instrument. Discard if turbid or if particulate matter is visible.

VI. QUALITY CONTROL:

A. **Self-Test**: At least once per shift at the start of each shift that the system is in use, perform the PFA-100 "Self Test" from the "Maintenance Menu". Document on the appropriate log.

B. **New Lot Number of Reagent**: Per manufacturer's recommendations, controls must be run in duplicate on each instrument using a healthy donor from the established normal pool. The system will be considered under control if the mean closure time (CT) falls within the established reference range. If the mean CT is outside the reference range, repeat using a new healthy donor. If the mean CT's from both individuals are outside the reference range contact Technical Services. If the mean CT from the second individual is within the reference range, the platelet function status and medication history of the first individual should be suspected.

C. **New Shipment of Reagent**: New shipment may or may not be the same as current lot number. A normal

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control (from established normal pool or healthy donor) should be run one time on each instrument with each new shipment of cartridges received or whenever there is need to verify the performance of the PFA instrument.

D. **Normal Donor Control:** Normal donor control is performed weekly on active PFA 100.
NOTE: Individuals who are potential donors should be free from any medication known to affect platelet function. Performance of this test is also monitored through participation in CAP proficiency surveys or equivalent.

VII. PFA CORRELATION:

A. A CV of ≤15% on the same instrument is acceptable for a normal platelet function test.

B. A CV of <15% between instruments is acceptable for a normal platelet function test.

C. There is no established allowable CV for abnormal platelet function tests

VIII. PROCEDURE:

A. Remove test cartridges from refrigerator. Allow to come to room temperature for a minimum 15 minutes. After removal of the cartridges from the pouch, close the pouch by using the re-closeable seal. Cartridges may be at room temperature for up to 4h. If not used, cartridges may be returned to refrigerator. If at room temperature for more than 4h or if cartridges are exposed to heat, discard.

B. Collagen/Epinephrine cartridges will have one slot identifying them; Collagen/ADP cartridges will have 2 slots identifying them. You can place cartridges in either well (left or right) – instrument will identify cartridge type.

C. When handling cartridges, grasp by tab end opposite circular membrane area.

D. Open cartridges by carefully pulling back the foil covering the larger end. DO NOT touch area of large circle. If this area is disturbed, the membrane is punctured and test is invalid.

E. Carefully place the cartridge into the well. Snap it securely into place. The top of the cartridge should be flush with the top of the carousel.

F. Mix specimen by inverting tube gently 3-4 times. Do not shake. Do not place specimen on tube rocker.

G. Add at least 820-830 mcL well mixed whole blood to the smaller opening on top of each cartridge. Pipette blood slowly down side of opening. Pipetting too fast will create air space at the bottom or air bubbles!

H. On the instrument, select “Run”. The instrument will request identification of patient specimen. Enter patient order number. If the same patient specimen is in both cartridges, you need only enter number once and select “Run Duplicate”. The instrument will assign the same I.D. number to both cartridges.

I. The test will take approximately 7-10 minutes.

J. When testing is complete, check specimen for clot . If present, cancel test. Results will be printed out on tape.

K. Remove cassette, discard cartridges into suitable biohazard waste container.

L. Replace cassette.

M. Enter results in LIS

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IX. EXPECTED RESULTS:

Refer to Coagulation Tests: Reportable Limits and Normal / Therapeutic Values procedure.

X. REPORTABLE RANGES:

Refer to Coagulation Tests: Reportable Limits and Normal / Therapeutic Values procedure.

XI. INTERPRETATION OF RESULTS:

A. Results of the PFA-100 are reported by the instrument as Closure Time (CT) in seconds. The PFA-100 test provides an indication of platelet function. Closure Time above the laboratory established cut-off may indicate the need for further diagnostic testing. Results should always be evaluated in conjunction with clinical history, presentation, and other laboratory findings (e.g. CBC and platelet aggregation). In cases where PFA-100 results do not agree with the clinical assessment, additional tests should be performed.

B. If a test result has an error code message, for example, max test time, flow obstruction, or insufficient sample with a letter code after the result, see Attachment C for interpretations of the error codes.
NOTE1 :**Per pathologist, if you are aware that the patient's PLT count is <50,000bill/L, cancel the PFA.**
NOTE 2: **If unable to achieve closure time on samples with low hematocrit (<30), cancel PFA due to low hematocrit.**

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XII. INTERFERING SUBSTANCES:

A. Presence of hemolysis may interfere with test results:
   1. There may be a reduction in hematocrit (<35)
   2. There may be release of ADP.

B. Certain fatty acids and lipids found in various human diets are widely known to inhibit platelet function for which the PFA-100 system was designed to detect. Neutral lipids, (e.g. cholesterol) generally have no effect on platelet function.

C. Platelet inhibiting agents directly affect platelet function.

XIII. LIMITATION:

A. Microthrombi in the sample or particulates introduced into the sample from the environment may adversely affect the test results.

B. Samples exhibiting high sedimentation properties should be run as single tests. (Hemodynamic properties of the sample may be altered, potentially affecting the result.)

C. The medication history of the patient should be reviewed, since many medications are known to affect platelet function.

D. Results will be affected by patients with a low hematocrit (<35%) or decreased platelet counts (<150,000 bill/L).

E. Certain fatty acids and lipids found in various human diets are known to inhibit platelet function. Physicians may wish to advise patients to refrain from fatty foods prior to testing.

XIV. REFERENCES:

A. PFA-100 Operating Manual. DADE International Inc., Miami FL 33172

B. DADE Behring PFA-100 Reagents product insert. DADE International Inc., Miami FL 33172 November 1998.

C. PFA-100 Analyzer Getting Started/Training Guide. DADE BEHRING 2002, Lit Number H634

Attachments

Attachment A-PFA MAINTENANCE.docx.pdf
Attachment C- PFA Guide to Common Status Messages.pdf
Attachment B-PFA Entering Lot Numbers.pdf

Approval Signatures

Step Description	Approver	Date
CP Chief Medical Director	Peter Millward: Chief, Pathology Service Line	5/13/2021
Coagulation Medical Director Designee	Marc Smith: System Med Dir, Coagulation	5/13/2021
Policy and Forms Steering Committee Approval (if needed)	Tamara Sabih: Medical Technologist Lead	5/11/2021
Policy and Forms Steering Committee Approval (if needed)	Gail Juleff: Project Mgr Policy	5/11/2021
System Manager	Rebecca Bacarella: Mgr Laboratory	5/11/2021
	Tamara Sabih: Medical Technologist Lead	5/4/2021

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