SIEMENS Healthineers

Dade® PFA Collagen/EPI Test Cartridge

COLL EPI CARTRIDGE

Dade® PFA Collagen/ADP Test Cartridge

COLL ADP CARTRIDGE

CE 0197

Revision bar indicates update to previous version.

Intended Use

COLL EPI CARTRIDGE, COLL ADP CARTRIDGE are in vitro diagnostic reagents for the quantitative determination of the collagen/epinephrine- and collagen/ADP-induced PFA closure times. The assays can be used as aid to diagnosis and follow up of congenital, acquired or therapy induced defects of primary hemostasis, including von Willebrand factor (vWF) dependent deficiencies, in patients at risk for or suspected of impaired primary hemostasis. The assays are performed using human sodium citrated whole blood by means of an automated, platelet adhesion and aggregation method under flow conditions.

For the determination of whole blood platelet adhesion and aggregation under flow conditions no reference preparation or method is available.

Summary and Explanation

The PFA-100® System and the INNOVANCE® PFA-200 System (PFA Systems) consist of an instrument and test cartridges in which the process of primary hemostasis with platelet adhesion and aggregation under flow conditions following a vascular injury is simulated in-vitro. Platelet-dependent hemostasis dysfunctions detected by the PFA Systems may be acquired, inherited, or induced by platelet inhibiting agents[1,2]. The most common causes of platelet-dependent hemostasis dysfunctions are related to uremia, von Willebrand disease (VWD), and exposure to drugs, such as acetylsalicylic acid (ASA).

The PFA Systems determine the time from the start of the test until the platelet plug occludes the defined aperture, and report that time interval as the Closure Time (CT). The CT is an indicator of platelet function in the analyzed whole blood sample. Platelet plug formation in the PFA Systems is affected by low platelet counts and/or activity, a reduced plasma level of von Willebrand factor and additionally, by a decreased hematocrit due to the flow process[3].

The COLL EPI CARTRIDGE (Col/EPI) is the primary test cartridge used to detect platelet-dependent dysfunction induced by intrinsic platelet defects, VWD or exposure to platelet inhibiting agents. The COLL ADP CARTRIDGE (Col/ADP) is used to indicate if the abnormal result obtained with the Col/EPI Test Cartridge may be caused by the effect of ASA or medications containing ASA.

During a vessel injury, the damaged blood vessels expose sub-endothelial collagen strands, which binds von Willebrand Factor (vWF) under high shear stress flow condition. Collagen-bound vWF subsequently binds platelets promoting the adherence of platelets to the side of injury, representing the first phase of primary hemostasis. At vascular lesions high amounts of ADP are released into the blood, while systemic reactions increase the level of epinephrine. All these substances, including

collagen, activate platelets through interaction with specific receptors, thereby amplifying the primary hemostasis process.

The PFA System in combination with Col/EPI and/or Col/ADP can be used for testing for primary hemostasis defects associated with an increased bleeding risk in subjects of 1 year and older[4–6].

The use of a standardized questionnaire in combination with the COLL EPI CARTRIDGE provided a high detection rate of impaired hemostasis in a pre-surgical screening approach including more than 5 000 patients scheduled for elective surgery. The vast majority of hemostasis defects identified affected the primary hemostasis; 98 % of all hemostasis defects were identified by the COLL EPI CARTRIDGE[7]. Corrective measures applied on basis of the screening and further specific test results resulted in pre-surgical correction of impaired primary hemostasis in most patients, and a reduction in total blood transfusions[8].

The COLL EPI CARTRIDGE is sensitive to ASA induced platelet inhibition, while the COLL ADP CARTRIDGE is not, which allows the differentiation between an ASA induced or non-ASA induced platelet dysfunction in case of a prolonged Col/Epi closure time. The Col/Epi closure time reflects the responsiveness to ASA therapy, and patients under ASA therapy with a shorter Col/Epi closure time were more likely to have vascular thrombotic events[9].

The COLL EPI CARTRIDGE and COLL ADP CARTRIDGE for the PFA System show a high sensitivity for the detection of reduced von Willebrand factor (vWF) activity; in a study including 213 genetically confirmed von Willebrand disease (VWD) in more than 4 000 patients screened over 16 years, sensitivity and negative predictive value both were 98 %[10]. Normal C/Epi plus normal C/ADP is considered inconsistent with type 2A, 2B, 2M and 3 VWD, and unlikely for type 1 VWD with levels of vWF below 25 to 30 %[11]. Furthermore, prolonged PFA closure times due to low vWF activity respond with shortening to therapy with desmopressin and vWF supplementation therapy[12].

The PFA closure time, especially with the COLL ADP CARTRIDGE, has proven to be sensitive to defects in high-molecular-weight multimers of vWF, occurring in acquired von Willebrand syndrome (aVWS), typically seen in patients with need for transcatheter aortic valve replacement (TAVR)[13]. In TAVR procedures paravalvular regurgitation (PVR) remains a relatively frequent and deleterious complication, and identification and grading of PVR in the catheterization laboratory remain an important and challenging clinical issue. A prolonged Col/ADP closure time > 180 seconds was highly predictive of persistent PVR after TAVR and major bleeding complication[14]. The PFA Col/ADP closure time allows to detect and monitor PVR in real-time, with an excellent negative predictive value. Testing performed directly in the catheterization laboratory improved the diagnosis of PVR and helps to rationalize the decision of whether or not to perform corrective measures[15].

Principles of the Procedure

The PFA Systems allow for rapid evaluation of platelet function on small samples of citrated whole blood based on work described by Kratzer and Born[16,17]. The single use PFA Test Cartridges consist of a number of integrated parts including a capillary, a sample reservoir and a biochemically active membrane with a central circular aperture. Citrated whole blood is aspirated from the sample reservoir through the capillary and the aperture, which expose platelets to high shear flow conditions. The membrane is coated with collagen, a subendothelial protein generally believed to be the initial matrix for platelet attachment. The attachment of platelets to collagen is thought to trigger the initial physiologic stimulus for platelet activation. In addition, the membrane is coated with either epinephrine or ADP, which are other physiologic agonists that, along with collagen, are widely used to activate platelets in aggregometry testing. At the beginning of a PFA test, Trigger Solution is dispensed to wet the membrane. During the test, platelets adhere to the collagen-coated membrane. Then, similar to aggregometry[18], platelets become activated and release their granule contents upon contacting agonists such as ADP or epinephrine. The release of granule contents is followed by adherence of platelets to each other to form aggregates. As a measure of platelet function in the PFA Systems, the process of platelet aggregation builds a platelet thrombus at the aperture thereby gradually diminishing and finally arresting the blood flow[3]. In optical aggregometry, platelet function is assessed by aggregate formation detected by changes in light transmittance.

The PFA Systems determine the time from the start of the test until the platelet plug occludes the aperture, and report that time interval as the Closure Time (CT). The CT is an indicator of platelet function in the analyzed whole blood sample. Platelet plug formation in the PFA Systems is affected by low platelet counts and/or activity, a reduced plasma level of von Willebrand factor, and additionally by, a decreased hematocrit because of the flow process[3].

Reagents

Reagent	Description	Storage	Stability
Dade® PFA Collagen/EPI Test Cartridge COLL EPI CARTRIDGE	Test cartridge unit containing a membrane coated with: • Type I Collagen, equine (2 µg) • Epinephrine bitartrate (10 µg)	2–25 °C May be used up to the expiry date indicated on the label if stored unopened.	2–25 °C: once opened, 3 months[a]; 15–25 °C: once opened, 4 hours[b]
Dade® PFA Collagen/ADP Test Cartridge COLL ADP CARTRIDGE	Test cartridge unit containing a membrane coated with: • Type I Collagen, equine (2 µg) • Adenosine-5'-diphosphate (50 µg)	2–25 °C May be used up to the expiry date indicated on the label if stored unopened.	2–25 °C: once opened, 3 months[a] 15–25 °C: once opened, 4 hours[b]

[a] in reclosed pouch
[b] in open pouch or outside the pouch

Warnings and Precautions

For *in-vitro* diagnostic use only.

For laboratory professional use.

According to EU regulation 2017/746, any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the EU Member State in which the user and/or patient is established.

Safety data sheets (MSDS/SDS) available on siemens-healthineers.com/sds.

Caution
COLL EPI CARTRIDGE, COLL ADP CARTRIDGE
This device contains material of animal origin and should be handled as a potential carrier and transmitter of disease.

All blood samples and blood components should be treated as potentially infectious. All samples should be handled in accordance with good laboratory practices using appropriate precautions.

Protective equipment should be worn when inserting or removing with whole blood loaded cartridges from the carousel.

Do not disassemble the test cartridge. There is a risk of exposure to blood droplets when removing the test cartridge from the carousel.

Dispose of hazardous or biologically contaminated materials according to the practices of your institution. Discard all materials in a safe and acceptable manner and in compliance with all government requirements.

Preparing Reagents

a. Allow the pouch containing the test cartridges to warm up to 15 to 25 °C prior to opening. This takes approximately 15 minutes. After removal of the cartridges, reseal the pouch.
b. Remove and discard the top foil from the test cartridges. This top foil only protects against dust and other particles. Performance and stability of the test cartridges are not affected in case of incomplete sealing by the top foil.
   Note: If the top foil has clearly been damaged or is missing, do not use the test cartridge. Discard it in an appropriate waste container.
c. Place the test cartridge(s) in the cassette of the PFA System and push until the test cartridge(s) securely snaps in place. (Refer to picture in the Introduction section of the respective Operating/ Instruction Manual).

Specimen Collection and Handling

Collecting the Specimen

All investigations of platelet function are strongly dependent on the correct method of blood collection. Venipuncture should be performed using a 21G or larger needle (20G or 19G). Blood should be drawn directly into an evacuated plastic or siliconized glass tube or syringe containing 0.11 mol/L or 0.13 mol/L (3.2 % or 3.8 %) **buffered** sodium citrate (1 part anticoagulant to 9 parts blood). Use of unbuffered sodium citrate anticoagulant is not recommended.

Important! After sample collection, ensure proper mixing of anticoagulant by gently inverting the tube by hand 3 to 4 times. Discard the sample if there is a venous collapse or stoppage of blood flow during collection. Do not use hemolytic blood samples. Samples must be stored undisturbed at 15 to 25 °C and are stable for 4 hours. For the COLL EPI CARTRIDGE, it is recommended that testing not be performed until 10 minutes after blood collection. **The collection method (both citrate concentration and venipuncture method) should be kept consistent.**

Procedure

Materials Provided

REF	Contents	
B4170-20	Dade® PFA Collagen/EPI Test Cartridge COLL EPI CARTRIDGE	20 × COLL EPI CARTRIDGE
B4170-21	Dade® PFA Collagen/ADP Test Cartridge COLL ADP CARTRIDGE	20 × COLL ADP CARTRIDGE

Note: The reagents are sold separately.

Materials Required but not Provided

Item	Description
REF B4170-50	PFA TRIGGER, Dade® PFA Trigger Solution
–	Evacuated blood collection tubes or syringes containing 3.8 % (0.129 M) or 3.2 % (0.105 M) buffered sodium citrate (1 part anticoagulant to 9 parts blood).
–	Pipetting devices capable of measuring up to 800 µL.
Coagulation analyzers[c], such as:	• PFA-100® System • INNOVANCE® PFA-200 System

[c] Availability of analyzers may vary by country.

Please note that the applications on other analyzers can be validated by the instrument manufacturer in accordance with the requirements of the REGULATION (EU) 2017/746 under their responsibility as long as the intended purpose and performance are not modified.

For details refer to the respective PFA Systems Operating/Instruction Manual.

No relevant differences were observed between singlicate and duplicate testing with respect to the reference intervals, diagnostic sensitivity and specificity, as well as diagnostic accuracy of the results. Therefore singlicate as well as duplicate testing can be performed with the PFA Systems. For information on singlicate testing, refer to the "Clinical Performance", page 9 section of these Instructions for Use.

When performing duplicate testing, the mean of the test results (including discrepant results, one above and one below the laboratory's established reference range) should be reported unless:

1. a non-closure was obtained at one position and at the other position a measurable CT within the reference range or
2. CT of one test position was higher than, or equal to, two times (2x) the value of the other position.

Samples with replicate test values meeting one of these criteria should be repeated with the same sample, if it is not older than four hours calculated from venipuncture. If the sample is older, the test must be repeated with a freshly drawn sample. Laboratories may wish to establish further, individual

criteria for repeat testing. If repeat testing does not resolve the discrepancy, contact Siemens Healthineers for assistance.

Sample loading

The following steps must be performed in sequence without interruption:

a. Mix the blood sample by inverting the collection tube gently by hand 3 - 4 times. Holding the cassette with test cartridge(s) on a flat surface, pipette 800 µL of blood into the smaller opening (sample reservoir opening) of the test cartridge by dispensing slowly along one of the inside surfaces. This will reduce the possibility of air entrapment in the sample reservoir.

Note: PFA Systems are incapable of detecting bubbles in the test cartridge.

b. Place the cassette with the test cartridge(s) into the incubation well(s) of the instrument so that the cassette is flush to the carousel surface. **Do not apply pressure to the sample reservoir opening**.

c. Start the test.

Disposal of used test cartridge(s)

Remove the cassette carefully from the carousel. Holding the cassette in one hand, remove test cartridge(s) by gently pulling the bottom of the cartridge(s) sideways until it unsnaps. Dispose of test cartridge(s) in a suitable biohazard waste container.

Internal Quality Control

System

At the start of each work shift, perform "Self Test" from the "Maintenance Menu". For details refer to the respective PFA Operating/Instruction Manual.

Cartridges

As part of the quality control it is recommended to test a control donor in duplicate with each new shipment of cartridges received or whenever the institution wishes to verify the performance of the system. The cartridges will be considered fully functional if the mean CT falls within the reference range established by the individual laboratory. If the mean CT is outside the reference range, repeat this procedure with a second individual from the laboratory's established control donor group. If the mean CT from both samples is outside the reference range, contact Siemens Healthineers. 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.

For the purpose of QC testing it is recommended to establish a control donor group. The CT of qualified QC donors should reveal an acceptable repeatability and values near the middle of the reference range of the laboratory.

The following procedure is an example of how to establish a control donor group:

1. Each donor should have no known diseases or medications influencing platelet function.
2. Test each potential donor by performing two replicates with COLL EPI CARTRIDGE only.
3. Qualify the donor if the duplicate mean is within 102 to 146 seconds and the CV is less than or equal to 15 %. (Note: This range was determined from the mean CT ±22 seconds of blood samples collected in 3.8 % buffered sodium citrate from 127 healthy blood donors in a Germany-based multicenter study, see also "Expected Values", page 6.

   Note: The acceptable range may need to be modified depending on the mean CT established by individual laboratories for healthy adults.

It is recommended that the laboratory run the quality control procedure in a manner consistent with its established quality control program and in conformance with local, state, and/or federal regulations or accreditation requirements.

Results

The result of a PFA test is reported as CT in seconds (s). The PFA test provides an indication of platelet function. A CT above the laboratory's established reference range may indicate the need for further diagnostic testing. Results should always be evaluated in conjunction with clinical history, clinical presentation and other laboratory findings (such as bleeding time, complete blood count (CBC) and platelet aggregometry).

In cases where PFA results do not agree with the clinical assessment, additional tests should be performed. The following pattern is expected when performing a PFA test on normal subjects respectively subjects with platelet dysfunction:

	Normal (n = 176)	ASA[d] (n = 120)	VWD (n = 28)	Glanzmann thrombasthenia (n = 4)
Col/EPI	normal	abnormal	abnormal	abnormal
Col/ADP	normal	normal	abnormal	abnormal

[d] See table under "ASA-induced Platelet Dysfunction", page 11.

Limitations

1. Microthrombi in the sample or particulates introduced into the sample from the environment could adversely affect the test results and/or cause a cancellation of the test due to the detection of a flow obstruction.
2. Blood samples with high sedimentation properties may experience some settling in position B while waiting to be tested in sequence with position A. Should settling occur, the hemodynamic properties of the sample may be altered, potentially affecting the result. Thus, it is recommended that samples exhibiting high sedimentation properties be run as single tests. In order to obtain duplicate measurements, single test should be performed in two separate runs.
3. Many medications are known to affect platelet function. Therefore, the medication history of the patient should be reviewed.
4. Only a CT above the laboratory's established reference range could reflect reduced platelet function caused by abnormally low hematocrit levels (< 35 %) and/or abnormally low platelet counts (< 150 000/µL). Specimens with hematocrit levels > 50 % or platelet counts > 500 000/µL have not been evaluated[3].
5. Certain fatty acids and lipids found in various human diets are known to inhibit platelet function and physicians may wish to advise patients to refrain from fatty foods prior to testing.
6. The performance characteristics of the PFA Systems have not been established in neonates and children under 1 year of age.
7. The performance characteristics of COLL EPI CARTRIDGE and COLL ADP CARTRIDGE have not been established for platelet inhibiting agents other than ASA.
8. In pathological samples with borderline platelet dysfunction caused by ASA drugs, highly variable results might be encountered during multiple testing.
9. Siemens Healthineers has validated use of these reagents on various analyzers to optimize product performance and meet product specifications. Please note that the applications on other analyzers can be validated by the instrument manufacturer in accordance with the requirements of the REGULATION (EU) 2017/746 under their responsibility as long as the intended purpose and performance are not modified. User defined modifications are not supported by Siemens Healthineers as they may affect performance of the system and assay results. It is the responsibility of the user to validate modifications to these instructions or use of the reagents on analyzers other than those included in Siemens Healthineers Application Sheets or these Instructions for Use.
10. Results of this test should always be interpreted in conjunction with the patient's medical history, clinical presentation and other findings.

Expected Values

The establishment of reference ranges for the cartridge types COLL EPI CARTRIDGE and COLL ADP CARTRIDGE with specimen collected in 3.8 % respectively 3.2 % buffered sodium citrate has been performed on 127 respectively 309[22] ostensibly healthy individuals with no previous history or laboratory results indicative of platelet dysfunction induced by intrinsic platelet defects, VWD or exposure to platelet inhibiting agents.

Anticoagulant	n	Cartridge Type	Mean[e] [s]	Reference Ranges[e] (5th–95th percentile) [s]
3.8 % (0.129 M) sodium citrate	127	Col/EPI	124	84 – 160
		Col/ADP	89	68 – 121
3.2 % (0.105 M) sodium citrate	309	Col/EPI	110	82 – 150
		Col/ADP	78	62 – 100

[e] based on duplicate determinations

As differences in donor population and other factors may affect results, it is recommended that each laboratory demonstrates transference of the reference ranges shown above (refer to CLSI guideline EP28 A3c[23]). If transference of these reference range cannot be verified, the laboratory should establish its own reference ranges.

Performance Characteristics

Precision

A study was conducted to characterize the variability of the PFA Systems. The samples used in this study were collected in evacuated tubes containing 3.8 % (0.129 M) buffered sodium citrate.

The study was performed on three cartridge lots for each type of cartridge. Blood samples were collected from four subjects with normal platelet function on three separate days over a period of eight days. Six replicates from each subject were tested on each day with the three cartridge lots of both types of cartridges.

The repeatability represents the variability in the results as derived from one healthy subject, from one draw, tested on one system, and with one lot of either COLL EPI CARTRIDGE or COLL ADP CARTRIDGE.

On top of repeatability, the total precision CV includes the lot-to-lot variability. However, total precision CV does not include the variability associated with the various subjects and samples, because such variability is not attributable to the assay.

The following results were obtained:

Col/ADP	Repeatability CV [%]	Total CV [%]
PFA-100®	8.95	9.02
INNOVANCE® PFA-200	9.54	9.54

Col/EPI	Repeatability CV [%]	Total CV
PFA-100®	10.51	10.52
INNOVANCE® PFA-200	9.35	9.48

In addition, data collected during a multicenter study was analyzed for variability due to test position. This data represented results from 176 subjects with normal platelet function tested in positions A and B. The duplicate CV was calculated at 13.5 % for COLL EPI CARTRIDGE and 10.0 % for COLL ADP CARTRIDGE. Results by site ranged from 10.4 % to 17.4 % with COLL EPI CARTRIDGE and from 7.7 % to 11.2 % with COLL ADP CARTRIDGE. For samples with CT near the upper reference range limit, the duplicate CV was 13.7 % for COLL EPI CARTRIDGE and 10.0 % for COLL ADP CARTRIDGE.

Interference

Interfering Substances

1. Hemolysis may interfere with test results. The lysis of red cells indicated by the presence of free hemoglobin could affect the PFA CT for two reasons:

   1) reduction in hematocrit and

   2) release of ADP.

Therefore, use of hemolyzed blood for PFA testing is not recommended.

2. Certain fatty acids and lipids found in various human diets are widely known to inhibit platelet function[19,20], for which the PFA System was designed to detect. Neutral lipids, such as cholesterol, generally have no effect on platelet function[21].
3. Commonly used drugs or certain substances in food may influence the closure time of the COLL EPI CARTRIDGE and/or COLL ADP CARTRIDGE.

The table below summarizes substances which affect the closure time of the COLL EPI CARTRIDGE and/or COLL ADP CARTRIDGE at the given and higher concentrations (according to an internal study). The results of the spiked sample and the native sample (same donation and position in the system) were evaluated using a paired t-test. A p-value < 0.05 was defined as significant interference.

Drug Category	Substance	Concentration	Concentration (S.I. units)	Influence on closure time	
				Col/EPI	Col/ADP
Antibiotic	Penicillin G	10 000 IU/mL	10^7 IU/L	prolongation	no influence
Analgesic	Ibuprofen	5 µg/mL	24.2 µmol/L	prolongation	no influence
Thrombolytic agent	Streptokinase	100 IU/mL	100 000 IU/L	prolongation	prolongation
Anti-platelet drug	Cilostazol	5 µg/mL	13.5 µmol/L	prolongation	no influence
	Tirofiban	0.1 µg/mL	0.2 µmol/L	prolongation	prolongation

Additional drugs that have influence on the closure time of the COLL EPI CARTRIDGE and/or COLL ADP CARTRIDGE as described in the literature are listed in the following table:

Drug Category	Substance	Influence on closure time		Reference
		Col/EPI	Col/ADP	
Anesthetic	Propofol[f]	prolongation	none	14
	Ropivacaine hydrochloride[g]	prolongation	prolongation	15
Hemostatic	Desmopressin (DDAVP)[h]	shortening	shortening	16
Analgesic	Diclofenac[h]	prolongation	n.d.[i]	17
	Ketorolac[h]	prolongation	n.d.[i]	17
	Indometacin[h]	prolongation	n.d.[i]	18
	Meloxicam[h]	prolongation	n.d.[i]	18
	Nabumeton[h]	prolongation	n.d.[i]	18
Plasma expander	Hydroxyethyl starch	prolongation	prolongation	19
Anti-platelet drug	Abciximab[h]	prolongation	prolongation	20
	Eptifibatid[h]	prolongation	prolongation	20
Vasodilator	Prostacyclin[h]	prolongation	prolongation	21
	Iloprost[h]	prolongation	prolongation	22

[f] Plasma concentration: 20 µg/mL
[g] Plasma concentration: 1.88 mg/mL
[h] *Ex vivo* measurement following administration of therapeutic doses
[i] not determined

Non-Interfering Substances

The following substances and drugs do not affect the closure time of the COLL EPI CARTRIDGE and COLL ADP CARTRIDGE when present in plasma at the concentrations indicated according to an internal

study. When evaluating samples spiked with the respective substance and the native samples (same donation and position in the system) using a paired t-test, the p-value was >0.05.

Drug Category	Substance	Concentration	Concentration (S.I. units)
ACE inhibitor	Captopril	25 µg/mL	115.1 µmol/L
Alcohol	Ethanol	5 µL/mL	85.7 mmol/L
Analgesic	Acetaminophen	25 µg/mL	165.4 µmol/L
Antiarrhythmic agent	Lidocaine	25 µg/mL	106.7 µmol/L
Anticoagulant	Low molecular weight Heparin	1.5 IU/mL	1 500 IU/L
Antidepressant	Fluoxetine	25 µg/mL	80.8 µmol/L
Antioxidant	Catechin	25 µg/mL	86.2 µmol/L
	α-Tocopherol	25 µg/mL	58.0 µmol/L
Beta-blocker	Propranolol	25 µg/mL	96.4 µmol/L
Bronchodilator	Theophyllin	25 µg/mL	138.8 µmol/L
Diuretic	Hydrochlorothiazid	25 µg/mL	84.0 µmol/L
Anti-inflammatory drug	5-Aminosalicylic acid	50 µmol/L	50.0 µmol/L
Glucocorticoid	Betamethason	25 µg/mL	63.7 µmol/L
Calcium channel blocker	Diltiazem	25 µg/mL	60.3 µmol/L
Coronar vasodilator	Nitroglycerin	0.1 µg/mL	0.4 µmol/L
Phosphodiesterase inhibitor	Caffeine	20 µg/mL	103.0 µmol/L
	Dipyridamol	10 µg/mL	19.8 µmol/L
Phosphodiesterase V inhibitor	Sildenafil	5 µg/mL	10.5 µmol/L
Statin	Pravastatin	25 µg/mL	58.9 µmol/L
Thyroid hormone	L-Thyroxine	0.4 µg/mL	0.5 µmol/L

Clinical Performance

A total of 328 specimens were tested in duplicate with a PFA System in a multicenter study. This population included samples from 176 subjects with normal platelet function and samples from 152 subjects with platelet dysfunction. A total of 115 subjects who were tested and enrolled in the normal platelet function group were tested post ASA ingestion and also enrolled in the platelet dysfunction group. The group of 176 normal subjects consisted of 61 % females and 39 % males with an age range between 18 and 57 years. The group of 152 platelet dysfunction subjects consisted of 66 % females and 34 % males with an age range between 16 and 66 years.

The Platelet Function Status (PFS) of each specimen was determined based upon results from a platelet function test panel (PT, APTT, platelet aggregometry, bleeding time, CBC [including platelet count], fibrinogen, Factor VIII:C, von Willebrand factor activity, von Willebrand factor antigen and multimer determination) and clinical history. Additional specimens for which platelet function test panel results could be interpreted differently by two individuals as being either normal or abnormal were classified as inconclusive for PFS. Samples with inconclusive PFS (n = 26) were excluded from the study.

Note: The only categories of platelet disorders studied with a PFA System included disorders that were ASA-induced, and disorders in patients with previous history or laboratory results indicative of platelet dysfunction induced by Glanzmann thrombasthenia or VWD. Patients on oral contraceptives, patients with coagulation factor abnormalities and patients with thrombocytopenia were not studied.

The abnormal group included specimens from subjects with (a) VWD (n = 28), (b) ASA-induced dysfunction (n = 120), and (c) Glanzmann thrombasthenia (n = 4). The PFA System clinical categorization for this study was based on a designated cut-off of 170 seconds. This cut-off was established with data from a previous study taking into account the overlap of CTs between the normal and abnormal populations. A concordance table for the PFA System categorization against platelet function status for the 328 specimens is presented below:

PFA System vs. Platelet Function Status (PFS) Comparison

	PFS Normal [n]	PFS Abnormal [n]
Col/EPI PFA Normal	156	6
Col/EPI PFA Abnormal	20	146

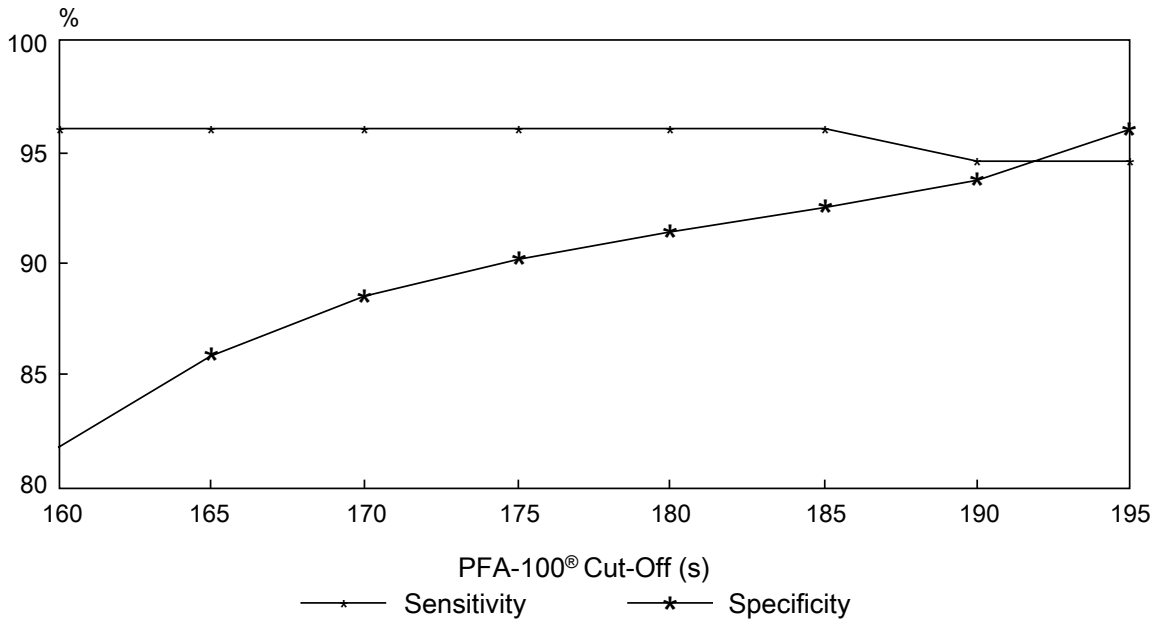
Overall diagnostic sensitivity and specificity were calculated from the table above at 96.1 and 88.6 %, respectively. Also, results were computed for each individual site and ranged from 86.7 to 100.0 % for diagnostic sensitivity and from 79.3 to 100.0 % for diagnostic specificity.

In addition to the calculation from the mean of the duplicate determinations, diagnostic sensitivity, specificity and accuracy for the PFA System were computed from the single determinations. The table below presents the results of these calculations. These results demonstrate that the clinical performance of the PFA System is not compromised by singlicate testing.

	Diagnostic Sensitivity [%]	Diagnostic Specificity [%]	Diagnostic Accuracy
Singlicate Determinations Position A	96.1	86.4	0.969
Position B	96.7	86.9	0.980
Duplicate Determinations	96.1	88.6	0.979
Confidence Limits	91.6–98.5	83.0–92.9	0.961–0.997

The data presented above reflects the clinical performance of a PFA System with COLL EPI CARTRIDGE using a cut-off at 170 seconds. The figure below depicts the observed diagnostic sensitivity and specificity when applying different cut-off values on the same study population. It is recommended that each laboratory establish its own cut-off based upon site population and internal procedure.

PFA System Sensitivity and Specificity at Different Clinical Cut-Offs



The concordance between a PFA System and aggregometry is shown in the table below. This table excluded 5 cases where aggregometry provided an inconclusive platelet function result. The overall proportion of agreement, expressed as a percentage, between a PFA System and aggregometry was calculated at 90.1 % from this table:

	Aggregometry	
	Normal [n]	**Abnormal [n]**
Col/EPI PFA Normal	145	12
Col/EPI PFA Abnormal	20	146

ASA-induced Platelet Dysfunction

Specimens drawn in 3.8 % (0.129 M) buffered sodium citrate from patients with normal platelet function following ASA ingestion were tested using COLL EPI CARTRIDGE in conjunction with COLL ADP CARTRIDGE on a PFA analyzer to evaluate platelet dysfunction due to ASA ingestion. A total of 120 specimens were tested in duplicate between 2 and 30 hours after ASA ingestion (325 mg). The results were as follows:

	Col/ADP Normal PFA CT ≤ 114 s [n]	**Col/ADP Abnormal PFA CT > 114 s [n]**
Col/EPI Normal CT ≤ 170 s	5	1
Col/EPI Abnormal CT > 170 s	87	27

The PFA System detected platelet dysfunction in 95 % of 120 ASA ingestion cases as indicated by the abnormal result obtained with the COLL EPI CARTRIDGE. The pattern of abnormal Col/EPI and normal Col/ADP was observed in 72.5 % of these 120 cases.

Differences in subject population, ASA dosage, the time of testing after ASA ingestion, and the anticoagulant used during blood sample collection, may produce results other than those listed.

Technical Assistance

For customer support, contact your local technical support provider or distributor.

siemens-healthineers.com

Applicable Version of electronic Instructions for Use

As Siemens Healthineers continuously monitors the product performance and safety, the users are required to ensure that they work with the correct revision of the instructions for the product lots in use. Please periodically review the availability of new electronic labeling revisions to ensure safe use of the product.

The IFU version number is visible on each product box label. Siemens Healthineers ensures that all products lots bearing the same IFU version number are compatible with the electronic labeling provided via siemens-healthineers.com/eIFU.

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Definition of Symbols

The following symbols may appear on the product labeling:

	Do not reuse		Use By
LOT	Batch Code	REF	Catalogue Number
	Caution		Manufacturer
EC REP	Authorized representative in the European Community		Contains sufficient for <n> tests
	Biological Risks	IVD	*In Vitro* Diagnostic Medical Device
	Temperature Limitation		Consult instruction for Use
	Non-sterile	CE	CE marking of conformity
CE 0197	CE marking of conformity with notified body ID number. Notified body ID number can vary.	CONTENTS	Contents
	Reconstitution volume	LEVEL	Level
	Keep away from sunlight and heat	WARNING	Warning
DANGER	Danger	RxOnly	Prescription device (US only)
UDI	Device Identification (UDI) barcode	REACH xx/xx/xx	REACH Authorization Number

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