D-DIMER-CHECK-1

Quantitative determination of D-Dimer in whole blood and citrated plasma samples

FOR EASY READER® AND EASY READER+® USE ONLY

Ref. 72091

I- PRINCIPLE

Fibrinogen is one of the main proteins of the blood coagulation system. As a result of the blood coagulation, thrombin is activating fibrinogen into fibrin monomers which are leading to clots formation (1). Fibrin clots are then digested by plasmin and D-Dimer, which is the main and the smallest component of fibrin clots lysis, is released into the bloodstream. The presence of D-Dimer in blood samples is an indicator of various coagulation disorders, including deep vein thrombosis (DVT), pulmonary embolism (PE) (2, 3) and atherosclerosis. D-Dimer assay is a widely used and simple exclusion method of DVT and PE (4) which is, in addition, not requiring any expensive laboratory instruments (5, 6).

In healthy individuals, D.Dimer concentration in blood is less than 400 ng /mL FEU (FEU: Fibrin Equivalent Unit).

The D-DIMER-CHECK-1 is a rapid screening test for the detection of D-Dimer in citrated plasma and whole blood samples.

Depending on the D-Dimer concentration, different lines will appear in the reading window, allowing the quantitative measurements of D-Dimer, when used in combination with the VEDALAB's rapid test readers.

II- D-DIMER-CHECK-1 KIT COMPONENTS

Each kit contains everything needed to perform 10 or 20 tests.

1- D-DIMER-CHECK-1 reaction devices	10	20
2- Disposable plastic pipettes	10	20
3- Diluent in dropper bottle	2.5mL	5mL
4- Instruction leaflet	1	1

5- **Controls (Optional): Positive control ref. V720 and Negative control ref. V721:** a freeze-dried preparation of a non-infectious compound in diluted human serum, tested and found negative for anti-HIV, anti-HCV and HBs antigen, containing 0.05 % sodium azide is optionally available as a positive and negative control (1x 0.25 mL). The concentration range is indicated on the vial label.

III- STORAGE AND STABILITY

1- All D-DIMER-CHECK-1 kit components, including optional control before reconstitution with distilled water, should be stored at any temperature between +4°C and +30°C in the sealed pouch.

2- **Do not freeze the test kit**.

3- The D-DIMER-CHECK-1 kit is stable until the expiry date stated on the package label.

IV- PRECAUTIONS

1- This test is designed for *in vitro* diagnostic use and professional use only.

2- Read the instructions carefully before using this test.

3- Handle all specimens as if they contain infectious agents. When the assay procedure is completed, dispose of specimens carefully after autoclaving them for at least one hour. Alternatively, they can be treated with 0.5% to 1% solution of sodium hypochlorite for one hour before disposal.

4- Wear protective clothing such as laboratory coats and disposable gloves while assaying samples.

5- Do not eat, drink or smoke in the area where specimens and kit reagents are handled.

6- Avoid any hands contact with eyes or nose during specimen collection and testing.

7- Do not use beyond the expiry date which appears on the package label.

8- Do not use a test from a damaged protective wrapper.

9- When the test is to be performed with whole blood, fresh samples should be used (>4 hours).

10- If plasma samples are to be assayed, please use only citrated plasma.

V- SPECIMEN COLLECTION AND PREPARATION

1- D-DIMER rapid test is performed on human citrated plasma or whole blood.

2- The specimen should be collected under the standard laboratory conditions (aseptically in such a way as to avoid haemolysis).

3- If anticoagulant is needed, only citrate should be used.

4- Each specimen should be treated as potentially infectious.

5- Whole blood samples should be tested immediately (< 4 hours). Finger prick samples should be assayed just after collection.

6- Due to the high fibrin concentration in the first volume of whole blood sample using the finger prick method, please discard it and collect a second sample of whole blood obtained from the same finger prick location to perform the test.

7- If the test is to be run within 48 hours after collection the specimen should be stored in the refrigerator (+2°C to +8°C). If testing is delayed more than 48 hours, the specimen should be frozen. The frozen specimen must be completely thawed, thoroughly mixed and brought to room temperature prior to testing. Avoid repeated freezing and thawing.

8- In case of cloudiness, high viscosity or presence of particulate matter into the plasma specimen, it should be diluted with equal volume (V/V) of diluting buffer (not provided but available upon request) before testing.

VI- ASSAY PROCEDURE

a) Controls testing

- Wait for 15 minutes after freeze-dried dissolving.
- Add the requested volume (25µL) with **lab pipette (disposable tips)** into the sample well of the cassette and proceed in the same way as for a patient's sample.
- The concentration range (**in ng/mL FEU**) is indicated on the vial label and obtained result must be within the specified range. The confidence range can change slightly depending on lot number.

- The reconstituted vial should be kept between +2°C and +8°C and should be used within 24 hours after reconstitution.

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b) Samples testing

Follow the below instructions or refer to the picture n°1.

1– Allow samples and D-DIMER-CHECK-1 test devices to come to room temperature prior to testing.

2– Remove the reaction device from its protective wrapper by tearing along the split.

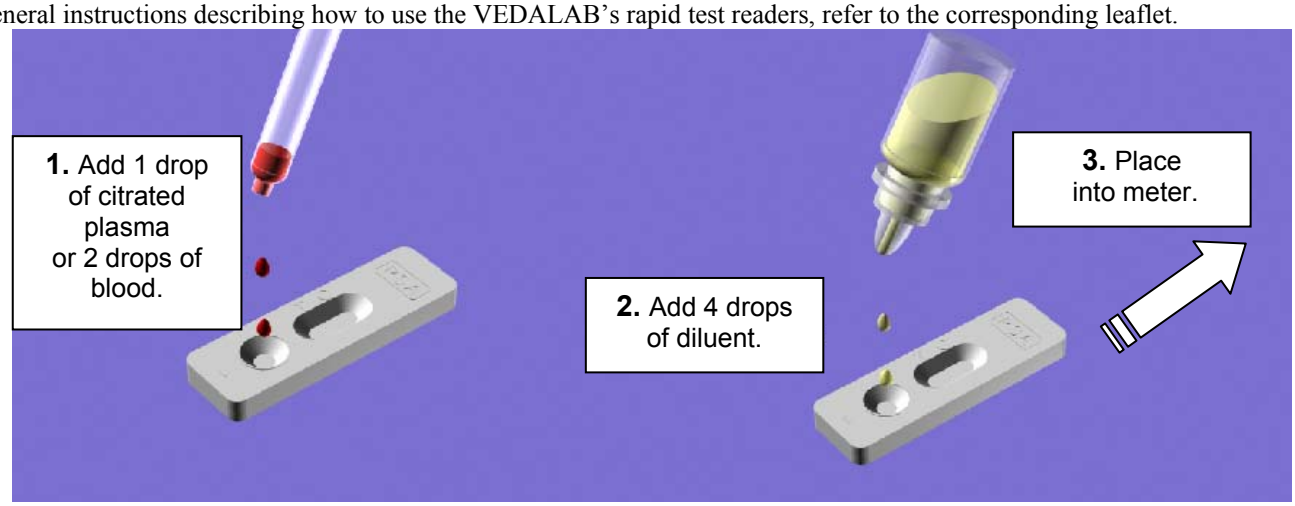
3– Label device with the patient's name or control number.

4– Fill the plastic pipette with sample or control and, by holding it vertically, dispense one drop (25 µL) of sample (citrated plasma) into sample well (▷). If the whole blood is used, dispense two drops (50 µL) into the sample well (▷) **and wait for the blood sample to be completely absorbed before adding diluent**.

5– Hold the diluent vial vertically and slowly add exactly 4 drops of diluent (150 µL) in the sample well (▷) **with an interval of 2-3 seconds between each drop.**

6– Read the result **(in ng/mL FEU**) after 15 minutes, either using the immediate or countdown reading mode (see corresponding leaflet).

For general instructions describing how to use the VEDALAB's rapid test readers, refer to the corresponding leaflet.



Picture n°1

VII- PERFORMANCES CHARACTERISTICS

a) Linearity

The measuring range is 250-5,000 ng/mL FEU.

For D-Dimer concentration lower than 250 ng/mL FEU, the result will be shown as "< 250 ng/mL FEU".

For D-Dimer concentration higher than 5,000 ng/mL FEU, the result will be shown as ">5,000 ng/mL FEU".

For samples whose concentration is higher than 5,000 ng/mL FEU, dilute with saline and repeat the assay as per instructions of Part. VI.

b) Accuracy

A study has been performed using plasma samples pre-assayed on VIDAS® (BIOMERIEUX) covering a range of 250 to 5070 ng/mL. Optical densities expressed as a function of D-Dimers concentrations are described by following linear curve:

$$Y = 0.0571x + 27.07 \text{ ng/mL } (r = 0.987).$$

The results show a good correlation ($r > 0.98$) of the values obtained with D-DIMER-CHECK-1 on VEDALAB's reader.

c) Sensitivity

Concentrations close to 200 ng/mL FEU are detected by D-DIMER-CHECK-1 test. In these cases, results will be rendered as "< 250 ng/mL FEU".

Levels higher than 400 ng/mL are generally considered as abnormal values.

d) Precision

A panel of 30 human plasmas pre-assayed on VIDAS ® analyser from BIOMERIEUX has been evaluated using the D-DIMER-CHECK-1 rapid test. Results are read with the VEDALAB's reader and reported in table I.

Four samples identified in bold typo are discrepant comparatively with the reference method.

Results obtained on samples n° 1, 7 and 26 by both methods indicate the same clinical diagnosis profile (positive).

Sample n°10 has a high level of CRP revealing a recent infection and the probable presence of poly-specific antibodies that could interfere with immunodiagnostic reagents (D-DIMER-CHECK-1 and VIDAS® BIOMERIEUX).

Negative, borderline and positive samples are correctly identified by the D-DIMER-CHECK-1 rapid test and correlation between the two methods attains 96.6 %.

Table I

Human plasmas identification	[D-DIMER] in ng/mL FEU Expected values (VIDAS BIOMERIEUX)	[D-DIMER] in ng/mL FEU Obtained values (D-DIMER-CHECK-1)
1	**2430**	**1851**
2	2380	2013
3	1130	1164
4	970	1094
5	540	570
6	3180	2724
7	**3090**	**2240**
8	8160	>5000
9	1350	1164
10	**150**	**706**
11	300	322

Human plasmas identification	[D-DIMER] in ng/mL FEU Expected values (VIDAS BIOMERIEUX)	[D-DIMER] in ng/mL FEU Obtained values (D-DIMER-CHECK-1)
12	340	337
13	540	595
14	560	509
15	3510	3336
16	1600	1485
17	1650	1545
18	2050	1900
19	2740	2481
20	5070	>5000
21	5640	>5000
22	990	963,3
23	1410	1510
24	1480	1204
25	3160	3540
26	**1380**	**1039**
27	250	300
28	510	600
29	920	1054
30	3787	4044

e) Hook effect

Plasma samples with a D-Dimer concentration of 10,000 ng/mL FEU were tested and showed consistently positive results.

f) Intra-assay reproducibility

Within run precision was evaluated by using 35 replicates of three commercially available references containing 893.34, 2132.03 and 4268.69 ng/mL FEU of D-Dimer as determined with quantitative D-DIMER-CHECK-1 for VEDALAB's reader.

The obtained CV (coefficient of variation) were respectively equal to 13.39%, 11.75% and 8.77%.

VIII- LIMITATIONS

1- The D-DIMER-CHECK-1 test is useful as a negative predictive diagnostic tool. A result lower than 400 ng/mL FEU excludes a DVT or PE.

2- A progressive increase of D-Dimer concentration is seen in patients over the age of 60 or during the second or third trimester of pregnancy (5). Therefore, the D-DIMER-CHECK-1 test should not be used to rule out DVT for these patients.

3- The D-Dimer assay should not be used for predicting DVT or PE for patients, suffering from cancer, experiencing an infectious or inflammatory process or having experienced a recent surgical act or trauma. A study performed among 255 patients hospitalized for pathologies other than venous thromboembolic disease showed a D-Dimer concentration exceeding 500µg/L is 78% of the patients (8).

4- As for any diagnostic procedure, the physician should evaluate the data obtained using this kit in the light of the other clinical information available.

5- If plasma sample are to be assayed use only citrated samples.

6- Use only fresh whole blood samples (< 4 hours) when test is performed with blood samples. Finger prick samples should be assayed just after collection.

7- Due to the high fibrin concentration in the first volume of whole blood sample using the finger prick method, please discard it and collect a second sample of whole blood obtained from the same finger prick location to perform the test.

8- This format of test is to be only used with VEDALAB rapid test readers (EASY READER® or EASY READER+®).

9- If the reading time (15 minutes) is not strictly respected, wrong results will be obtained.

10- This format of test should not be used for visual reading.

11- As for any diagnostic method or for any measurements through analysers, there is a variability of the obtained result. Therefore, a confidence range of +/- 25% should be considered for the final value and for the clinical significance of the result.

IX- BIBLIOGRAPHY

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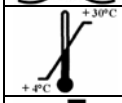
	Read the instructions before use		For *in vitro* diagnostic use
	Temperature limitations		Do not reuse
	Manufacturer		

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