FABP-CHECK-1

Quantitative determination of Heart-type fatty acid-binding protein in whole blood, serum or plasma samples

Ref. 98091 (20 tests) / Ref. 98091-10T (10 tests)

- FOR EASY READER® AND EASY READER +® USE ONLY –

I- INTENDED PURPOSE

FABP-CHECK-1 is a rapid screening test for the detection of human FABP (fatty acid-binding protein) in whole blood, serum or plasma samples to be used as a tool by medical healthcare professionals in assessing cardiac disorders such as acute myocardial infarction (AMI). The sole measurement of FABP concentration is not sufficient to diagnose myocardial ischemic damage. Additional clinical examinations such as echography/echocardiogram or electrocardiogram (ECG) are generally necessary to confirm the diagnosis.

II- PRINCIPLE

Heart-type fatty acid-binding protein (H-FABP or FABP) is a small cytosolic protein that is abundant in cardiac tissue. After myocardial ischemic damage, H-FABP can be detected in the blood as early as within 1 hour after onset of chest pain, with peak values reached at 3–6 hours and plasma levels returning to normal within 24–30 hours (1). The combination of initial H-FABP release after symptom onset, rapid kidney clearance from the circulation, and high cardiac specificity makes it as an early marker for the diagnosis of AMI (2,3). The H-FABP can also be used to detect perioperative AMIs and detect re-infarction if it occurs within 10 hours after symptom onset (4).
FABP-CHECK-1 is a diagnostic rapid test kit to be used if the occurrence of an acute myocardial infarction (AMI) is suspected. The test kit can be used between 30 minutes and 6 hours after onset of symptoms. FABP levels return to base line within 24 hours.

FABP-CHECK-1 is a rapid quantitative assay for the measurement of Heart-type fatty acid-binding protein in serum, plasma or whole blood samples. The method employs a unique combination of two different monoclonal dye conjugate and monoclonal solid phase antibodies to identify FABP in the test samples with a high degree of sensitivity. As the test sample flows through the absorbent device, the antibody-dye conjugate binds to the h-FABP forming an antibody-antigen complex. This complex binds to the anti FABP antibody in the reaction zone (T) and produces a pink-rose color band when FABP concentration is sufficient. The reaction mixture continues flowing through the absorbent device past the reactive zone and control zone (C). Unbound conjugate binds to the reagents in the control zone (C), producing a pink-rose color band, demonstrating that the reagents are functioning correctly.

III- FABP-CHECK-1 TEST KIT COMPONENTS

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

1- FABP-CHECK-1 reaction devices:	10	20
2- Disposable plastic pipettes:	10	20
3- Diluent in a dropper bottle containing saline buffer, detergent and sodium azide (NaN_3 < 0.1%):	2.5 mL	5 mL
4- Instruction leaflet:	1	1

5- **Positive and negative controls (ref. V9800 and ref. V9801) (optional)** 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 level is indicated on the vial label.

IV- STORAGE AND STABILITY

1- All FABP-CHECK-1 test components (except controls) should be stored at any temperature between +4°C and +30°C.
2. The controls should be stored between +2°C and +8°C before reconstitution.
2- Do not freeze the test kit.
3- FABP-CHECK-1 test is stable until the expiry date stated on the package label.

V- PRECAUTIONS

1- This test is designed for *in vitro* diagnostic use and professional use only.
2- Read carefully the instructions before using this test.
3- Handle all specimens as if they contained 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 contact between hands and 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.

VI- SPECIMEN COLLECTION AND PREPARATION

1- FABP-CHECK-1 is to be performed on human serum, heparinized/citrated/EDTA plasma or whole blood samples.
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, EDTA or heparin should be used.
4- Each specimen should be treated as if potentially infectious.
5- Whole blood samples should be tested immediately (< 4 hours). Finger prick samples should be assayed just after collection.
6- 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.
7- In case of cloudiness, high viscosity or presence of particulate matter into the serum specimen, it should be diluted with equal volume (V/V) of diluting buffer (not provided but available upon request) before testing.

CE

VII- ASSAY PROCEDURE

a) Control testing
- For both freeze-dried positive and negative controls, reconstitute each vial with 0.25 mL of distilled water.
- 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**) 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.

b) Sample testing

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

1– Allow samples and FABP-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 serum or 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 add exactly 4 drops (150 µL) of each diluent into the sample well (▷) **with an interval of 2-3 seconds between each drop.**
6– Read the result **(in ng/mL**) after 10 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.

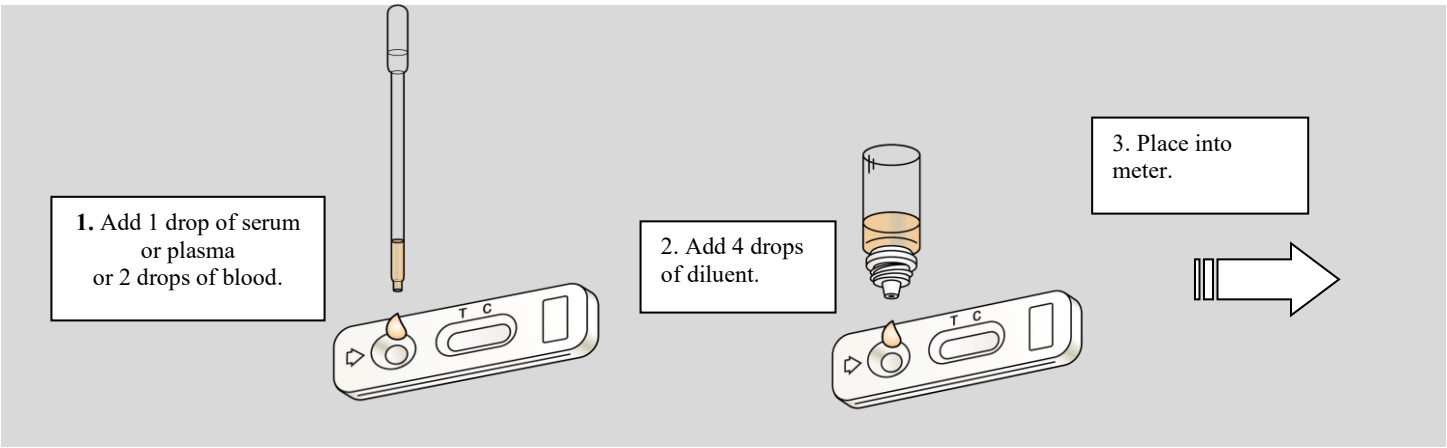


Figure n°1

VIII- PERFORMANCES CHARACTERISTICS

a) Linearity
A study has been performed using dilutions of FABP purified antigen in negative sample and covering a range of 0 to 120 ng/mL.
The dose response obtained with FABP-CHECK-1 test fits a linear regression: Y = 1.0176x+0.6129 (r = 0.99)

The measuring range is 2 - 120 ng/mL.
For FABP concentration below 2 ng/mL, the result will be given as " < 2 ng/mL".
For FABP concentration over 120 ng/mL, the result will be given as " > 120 ng/mL"
For samples whose concentration is higher than 120 ng/mL, dilute with saline and repeat the assay as per instructions of Part.VII.

b) Sensitivity
Concentrations close to 1 ng/mL are detected using FABP-CHECK-1 test. In these cases, results will be shown as "< 2 ng/mL".
Levels higher than 6 ng/mL are generally considered as abnormal values.

c) Accuracy
A comparative study was performed using 93 pre-assayed serum/plasma samples (HUMAN FABP3 kit) and FABP-CHECK-1 test. Obtained results show a correlation of 80.65% (CI 95% [69.76 – 88.72] *) between both methods (figure 2).

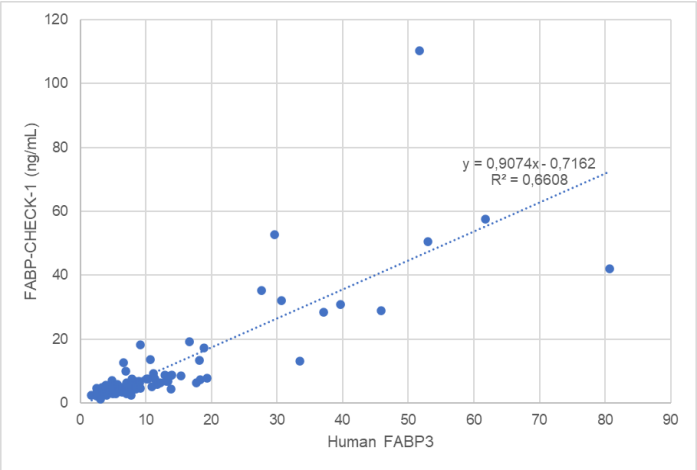


Figure n°2

The diagnostic specificity, sensitivity and correlation performances are calculated as shown in table 4.

Diagnostic sensitivity	(42/59) x100=71.19% [55.25-83.90] CI 95%*
Diagnostic specificity	(33/34) x100=97.06% [83.97-100] CI 95%*
Correlation	((42+33) /93) x100=80.65% [69.76-88.72] CI 95%*
Positive predictive value	(42/43) x100=97.67% [87.08-100] CI 95%*
Negative predictive value	(33/50) x100=66.00% [47.83-81.30] CI 95%*

Table 4: Diagnostic sensitivity and specificity

*CI 95%: 95% Confidence interval

d) Hook effect
No hook effect was observed up to a FABP concentration of 10,000 ng/mL (10 µg/mL). The reader result was: "> 120 ng/mL".

e) Intra-assay reproducibility
Within run precision was evaluated by using 25 replicates of three human samples obtained by serial dilution in negative serum sample of a purified FABP antigen. The three samples were containing respectively 10, 25 and 50 ng/mL of FABP as determined with quantitative FABP-CHECK-1 test.
The obtained CVs (coefficient of variation) were respectively equal to 8.8%, 11.7% and 7.4%.

f) Interferences
1- Bilirubin, triglycerides and hemoglobin
3 serum samples containing respectively 0; 10 and 75 ng/mL of FABP spiked with either bilirubin (30 mg/L), triglycerides (15 g/L) or hemoglobin (5 g/L) were triplicate tested using the FABP-CHECK-1 quantitative rapid test. The results show that bilirubin, triglycerides and hemoglobin do not interfere up to concentrations of 30 mg/L, 15 g/L and 5 g/L respectively.

2- Rheumatoïd factor (RF)
1 negative serum sample (<6 ng/mL) spiked with rheumatoid factor (475 IU/mL) was tested in triplicate using the FABP-CHECK-1 quantitative rapid test. The results show repeated measured values "<6 ng/mL" and therefore RF concentration up to 475 IU/mL do not interfere in the FABP-CHECK-1 quantitative rapid test.

3- HAMA
1 negative serum sample (<6 ng/mL) spiked with HAMA (Human anti-mouse antibodies) type 1 or 2 was tested in triplicate using the FABP-CHECK-1 quantitative rapid test. The results show that the HAMA of type 2 (appearing in patients after treatment with monoclonal antibodies) does not interfere with the FABP-CHECK-1 quantitative rapid test. HAMA of type 1 (antibodies that can be present in healthy patients) can interfere with the FABP-CHECK-1 quantitative rapid test at the tested concentrations levels.

4- Anticoagulants
No interference of heparin, citrate or EDTA was observed.

g) Matrix effect
30 serum or EDTA plasma samples spiked with red blood cells have been assayed using the FABP-CHECK-1 quantitative rapid test. There was no difference in the results, i.e., no matrix effect, observed between the plasma samples and the whole blood samples.

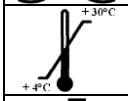
IX- LIMITATIONS

1- As for any diagnostic procedure, the physician should evaluate the data obtained using this test by other clinical methods.
2- If the testing is performed at the border of the diagnostic window (i.e., between 30 minutes and 24 hours after onset of symptoms), a false negative test result cannot be completely ruled out. Therefore, a negative result within the first hours of the onset of symptoms does not rule out AMI with certainty. If suspected, repeat the test at appropriate intervals.
3- A negative test result does not exclude the possibility that an acute myocardial infarction (AMI) has occurred.
4- H-FABP can be elevated in patients with renal insufficiency or *angina pectoris*. In low amounts, H-FABP is also present in skeletal muscle. Therefore, it can be elevated in individuals that performed physically prior to the testing.
5- High levels of RF (Rheumatoid factor) or CRP (C-reactive protein) may create interferences and therefore lead to false positive results.
6- The test is designed to eliminate the potential interference of human antibodies to murine IgG (HAMA). However high level of HAMA could give falsely positive results.
7- **Use only fresh whole blood samples (< 4 hours) when test is performed with blood samples. Finger prick samples should be assayed just after collection.**
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 (10 minutes) is not strictly respected, wrong results will be obtained.

10- This format of test should not be used for visual reading.
11- As it is true 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.

X- BIBLIOGRAPHY

1. Chan, Sanderson et al. A superior early myocardial infarction marker. Human heart-type fatty acid-binding protein. Z Kardiol. 93: 388-397. 2004.
2. Glatz, Van der Voort, Hermens. Fatty acid-binding protein as the earliest available plasma marker of acute myocardial injury. J. Clin. Ligand. Assay. 25: 167-177. 2002.
3. Nakata. Human heart-type fatty acid-binding protein as an early diagnostic and prognostic marker in acute coronary syndrome. Cardiology 99: 96–104. 2003.
4. Fransen et al. Perioperative myocardial tissue injury and the release of inflammatory mediators in coronary artery bypass graft patients. Cardiovasc. Res. 45: 853–859. 2000.

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

 Manufactured by VEDALAB – France

CHANGES DESCRIPTION
Changes type:

- N/A — Not Applicable (creation)
- Technical change — Addition, revision and/or removal of information related to the product.
- Administrative — Implementation of non-technical changes noticeable to the end-user.

Changes type	**Change description**
Technical	- Modification of VIII- c) accuracy - Addition in VIII- correlation, interferences, matrix effect
Administrative	Addition of I- Intended purpose and Ref 10 and 20 tests

Note: Minor typographical, grammar, spelling and formatting changes are not reported in the change details.