# TROPONIN I-CHECK-1

## Quantitative determination of Troponin I in whole blood, serum or plasma samples

## Ref. 28091 (20 tests) / Ref. 28091-10T (10 tests)

## FOR EASY READER® AND EASY READER+® USE ONLY

### I- INTENDED PURPOSE

The TROPONIN I -CHECK-1 is a rapid screening test for the detection of cardiac troponin I protein in whole blood, plasma or serum samples to be used as a tool by medical healthcare professionals assessing myocardial ischemia disorder such as acute myocardial infarction (AMI). The sole measurement of concentration or determination of troponin I is not sufficient to diagnose acute coronary syndrome or associated cardiac complications such as myocardial infarction.

### II- PRINCIPLE

Troponin I (TnI) is one of the thin filament-associated regulatory proteins of muscle (1). It is encoded by three different genes that are differentially expressed by the various muscle tissues, resulting in slow – and fast skeletal and cardiac TnI isoforms (2).

The unique amino acid sequence of cTnI makes it an ideal candidate for the laboratory detection of acute myocardial infarction (AMI) and has facilitated the development of monoclonal antibodies that do not cross-react with skeletal muscle troponins (3). Published studies from various groups have demonstrated the utility of cTnI measurement for detection of AMI (3, 4, 6, 7, 8).

CK-MB and cTnI both elevated beyond normal reference limits within 4-6 hours after infarction. Typical reference limits were, as reported by Bodor *et al* (3), 6.7 ng/mL for CK-MB and 3.1 ng/mL for cTnI.

Likewise, each report sites similar time frames for the peak values of CK-MB and cTnI : CK-MB peaked in 13-15 hours, cTnI in 11-15 hours. Typical ranges were 39 –185 ng/mL for CK-MB and 18.5 – 188 ng/mL for cTnI (5).

However, CK-MB level returns to normal after 36-48 hours, while levels of cTnI remains elevated for up to 6-10 days. The level of cTnI is very low in normal healthy people, and not detected in patients with skeletal muscle injury. Therefore, cTnI is a specific marker for diagnosis of AMI.

TROPONIN I-CHECK-1 is a rapid quantitative assay for the detection of cardiac Troponin I in serum, plasma or whole blood samples. The method employs a unique combination of monoclonal dye conjugate and polyclonal solid phase antibodies to identify troponin 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 troponin forming an antibody-antigen complex. This complex binds to the anti troponin antibody in the reaction zone (T) and produces a pink-rose colour band.

In the absence of troponin, there is no band in the reaction zone (T). The reaction mixture continues flowing through the absorbent device past the reactive zone (T) and control zone (C). Unbound conjugate binds to the reagents in the control zone (C), producing a pink-rose colour band, demonstrating that the reagents are functioning correctly.

### III. TROPONIN I-CHECK-1 KIT COMPONENTS

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

| | | |
|---|---|---|
| 1- TROPONIN I -CHECK-1 reaction devices: | 10 | 20 |
| 2- Disposable plastic pipettes: | 10 | 20 |
| 3- Diluent in a dropper bottle: | 2.5mL | 5mL |
| 4- Instruction leaflet: | 1 | 1 |

**5- Controls (Optional):**

**Positive control (ref. V280) and Negative control (ref. V281)**: 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.

### IV- STORAGE AND STABILITY

1- All TROPONIN I -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 TROPONIN I -CHECK-1 kit 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- TROPONIN I rapid test is performed on human serum, 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, 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.

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## VII- ASSAY PROCEDURE

**IMPORTANT: Switch the reader on and allow it to warm up for at least 30 minutes before performing any measurements.**

**a) Control testing**

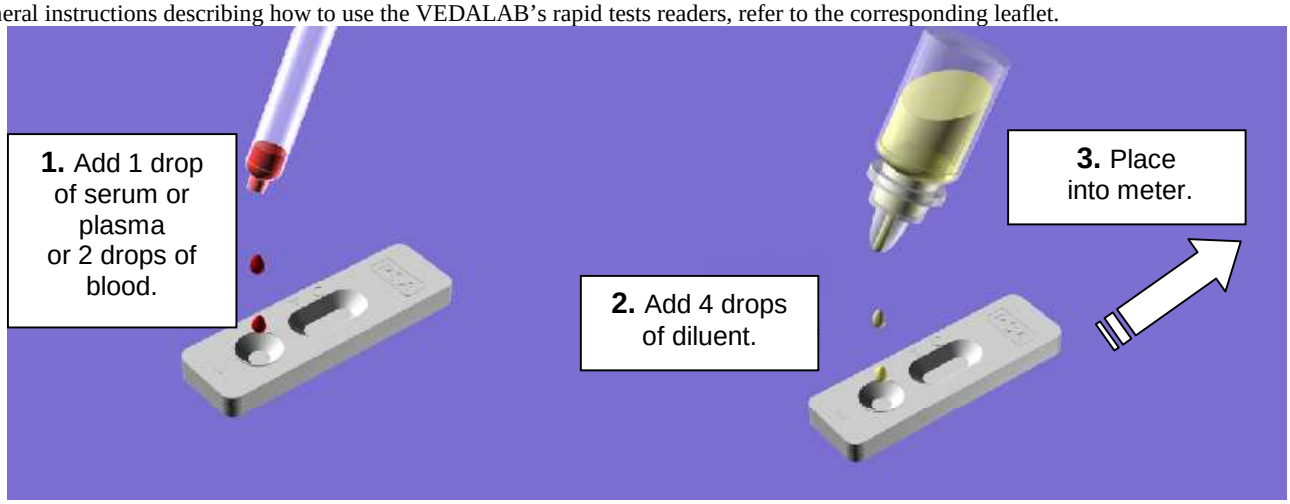
- Wait for 15 minutes after freeze-dried dissolving.
- Add the requested volume (25µL) with **lab pipette (disposable tips)** into the sample well of the cassette and proceed in the same way as for a patient's sample.
- The concentration range (**in ng/mL**) is indicated on the vial label and obtained 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 5 hours after reconstitution due to the very low stability of troponin I**.

**b) Sample testing**

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

1– Allow samples and TROPONIN I -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 fully absorbed before adding diluent**.
5– Hold the dropper bottle vertically and slowly add exactly 4 drops of diluent (150 µL) into the sample well (▷) **with an interval of 2-3 seconds between each drop.**
6– Read the result **(in ng/mL**) after 20 minutes, either using the immediate or countdown reading mode (see corresponding leaflet).

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




**Picture n°1**

## VIII- PERFORMANCES CHARACTERISTICS

**a) Linearity**

The linearity measuring range is 0-50 ng/mL.
For samples whose concentration is higher than 50 ng/mL, dilute with saline and repeat the assay as per instructions of Part. VI.

**b) Accuracy**

1- Standards

A study has been performed using five concentrations of DADE-BEHRING calibrators determined in duplicate on TROPONIN I-CHECK-1 rapid tests. The results show the good correlation of the values obtained with TROPONIN I-CHECK-1 on VEDALAB's reader.

Correlation figures : $y = 1.258\ x - 0.3435$ $r = 0.997$

2. Human samples

Another study has been performed using a panel of 80 human sera pre-assayed on BECKMAN ACCESS analyser. The decision value for AMI is respectively 0.8 ng/mL for TROPONIN I-CHECK-1 quantitative assay and 0.5 ng/mL for BECKMAN reagent. Samples with a concentration level higher than the decision value are identified as positive (+); samples with a concentration level lower than the decision value are identified as negative (-). The obtained results are shown in table 1.

Table 1

| Human sera identification | TROPONIN I-CHECK-1 | | *BECKMAN* | | Human sera identification | TROPONIN I-CHECK-1 | | *BECKMAN* | |
|---|---|---|---|---|---|---|---|---|---|
| | [TRP-I] in ng/mL | Result | [TRP-I] in ng/mL | Result | | [TRP-I] in ng/mL | Result | [TRP-I] in ng/mL | Result |
| 13 | 0.09 | - | 0.26 | - | 120 | 0.5 | - | 0.01 | - |
| **14** | **0.83** | **+ (d)** | **0.04** | - | 121 | 0.39 | - | 0.01 | - |
| 15 | 1.70 | + | 1.65 | + | 122 | 5.55 | + | 6.6 | + |
| 16 | 4.95 | + | 5.13 | + | 123 | 8.08 | + | 8.64 | + |
| 17 | 3.02 | + | 1.71 | + | 124 | 0.72 | - | 0.23 | - |
| 31 | 0.17 | - | 0.01 | - | 125 | 0.28 | - | 0.01 | - |
| 32 | 0.28 | - | 0.01 | - | 126 | 0.44 | - | 0.01 | - |
| 33 | 0.56 | - | 0.01 | - | 127 | 0.22 | - | 0.01 | - |
| 34 | 0.06 | - | 0.04 | - | 128 | 0.05 | - | 0.01 | - |
| 35 | 0.05 | - | 0.02 | - | 160 | 0.07 | - | 0.01 | - |
| 36 | 0.5 | - | 0.01 | - | 161 | 0.39 | - | 0.01 | - |
| 38 | 0.08 | - | 0.03 | - | 162 | 0.28 | - | 0.01 | - |
| **39** | **1.09** | **+ (d)** | **0.01** | - | 163 | 0.39 | - | 0.01 | - |
| 40 | 0.09 | - | 0.01 | - | 164 | 0.17 | - | 0.01 | - |
| 41 | 0.44 | - | 0.01 | - | 165 | 0.44 | - | 0.01 | - |
| 42 | 0.09 | - | 0.01 | - | 166 | 0.39 | - | 0.01 | - |
| 43 | 0.39 | - | 0.01 | - | 167 | 0.28 | - | 0.01 | - |
| 44 | 0.09 | - | 0.01 | - | 168 | 0.39 | - | 0.01 | - |
| 45 | 1.97 | + | 0.94 | + | **22** | **0.83** | **+(d)** | **0.06** | **-** |
| 46 | 0.04 | - | 0.01 | - | **23** | **0.89** | **+(d)** | **0.29** | **-** |
| 47 | 0.28 | - | 0.01 | - | **24** | **1.09** | **+(d)** | **0.07** | **-** |
| 48 | 0.22 | - | 0.01 | - | 25 | 3.38 | + | 2.72 | + |
| 49 | 0.17 | - | 0.01 | - | **26** | **0.56** | **-(d)** | **0.72** | **+** |
| 103 | 0.07 | - | 0.04 | - | 27 | 0.33 | - | 0.21 | - |
| 104 | 0.44 | - | 0.01 | - | 28 | 0.28 | - | 0.09 | - |
| 105 | 0.33 | - | 0.01 | - | 29 | 0.61 | - | 0.07 | - |
| 106 | 0.56 | - | 0.01 | - | 30 | 0.39 | - | 0.09 | - |
| 107 | 0.61 | - | 0.01 | - | 31 | 0.28 | - | 0.01 | - |
| 108 | 0.04 | - | 0.01 | - | 52 | 0.44 | - | 0.02 | - |
| 109 | 0.28 | - | 0.01 | - | 53 | 0.67 | - | 0.01 | - |
| 110 | 0.17 | - | 0.01 | - | 54 | 0.08 | - | 0.01 | - |
| 111 | 0.11 | - | 0.01 | - | 60 | 0.09 | - | 0.01 | - |
| 112 | 0.39 | - | 0.01 | - | 61 | 0.5 | - | 0.11 | - |
| 113 | 0.33 | - | 0.01 | - | 62 | 0.04 | - | 0.01 | - |
| 114 | 0.28 | - | 0.01 | - | 63 | 0.28 | - | 0.01 | - |
| 115 | 0.61 | - | 0.01 | - | 64 | 0.08 | - | 0.02 | - |
| 116 | 0.39 | - | 0.47 | - | 65 | 0.05 | - | 0.01 | - |
| 117 | 0.33 | - | 0.01 | - | 66 | 11.96 | + | 12.66 | + |
| 118 | 0.22 | - | 0.01 | - | 68 | 0.89 | + | 0.63 | + |
| 119 | 0.39 | - | 0.02 | - | 70 | 2.4 | + | 2.51 | + |

**(d): discrepant results.**

Discrepant results are obtained with six serum samples (**in bold**).
- Samples N° 14, 39, 22, 23 and 24: results obtained using TROPONIN I-CHECK-1 test are slightly over the decision level (0.8 ng/mL), involving no actual risk for patients.
In most cases (N° 14, 22, 23, 24, 26), the BECKMAN result is also over the AMI risk limit (0.04 ng/mL).
- Sample N°26: results obtained with this sample are close to the AMI cut-off in both cases, i.e. slightly lower for VEDALAB's reader or slightly higher for BECKMAN.
This indicates in all cases that further testing should be done later on, depending on the physiological status of the patient, as values indicate a possible AMI. An overall agreement of 92.5 % (CI* 95% [84.6 – 96.5] ) has been established with TROPONIN I-CHECK-1 and BECKMAN analyser.
*CI 95% : 95% confidence interval

**c) Sensitivity**
The detection limit is lower than 0.5 ng/mL. Any concentration around or higher than 0.8 ng/mL, for serum and plasma samples, or around or higher than 1.0 ng/mL for whole blood samples, may suggest a possible AMI case and further investigations should be made. Troponin I elevated levels are also observed in setting of systemic inflammatory response syndrome (SIRS), sepsis or septic shock (9).

**d) Specificity**
Negative sera assayed using the STRATUS-DADE analyser were found constantly negative using TROPONIN I-CHECK-1. No cross-reaction has been observed with the skeletal muscle Troponin I.

**e) Hook effect**
No hook effect has been observed up to 5 µg/mL both for complexed and purified form of Troponin I.

**f) Intra-assay reproducibility**
Within run reproducibility was evaluated by performing 26 replicates of three commercially available references containing 4.29, 17.33 and 36.87 ng/mL of TROPONIN I as determined with quantitative TROPONIN I-CHECK-1 for VEDALAB's reader.
The obtained CVs (coefficient of variation) were respectively equal to 9.91%, 7.55% and 9.45%.

**g) Inter-assay reproducibility**
Between run reproducibility was determined by performing four serum samples containing 1.7; 4.8; 10.1 and 16.2 ng/mL of TROPONIN I respectively measured using three different lots of TROPONIN I-CHECK-1 (3 replicates / lot). The obtained coefficients of variation (CV) are 12.7, 14.6; 13.6 and 10.7% respectively.

**h) Interferences**
1/ Hemoglobin, bilirubin and triglycerides
Negative (0.003ng/mL), weak positive (1.7ng/mL) and strong positive (7.56ng/mL) samples spiked with hemoglobin (final concentration : 5g/L), bilirubin (final concentration : 0.3g/L) of triglycerides (final concentration: 30g/L) did not show any effect on samples status (negative or positive).
2/ Anticoagulants
Plasma samples containing different anticoagulants were assayed indicating no matrix effect of citrate, EDTA and heparin.

**i) Cross reactions**
1/ Rheumatoid factor
30 serum samples containing high levels of rheumatoid factor (RF) have been assayed with the TROPONIN I-CHECK-1 test. No false positive result was observed in presence of RF and therefore there was no interference of RF in TROPONIN I-CHECK-1 test.
2/ HAMA
Positive samples in HAMA (human anti-mouse antibodies) type 1 and type 2 and negative Troponin I samples have been tested. No interference was observed for HAMA type 2. Weakly false positive results can be observed with high concentrated HAMA type 1. HAMA type 2 are found in persons treated by immunotherapy while HAMA type 1 are present in healthy persons.

## IX- LIMITATIONS
1- As for any diagnostic procedure, the physician should confirm the data obtained using this test by other clinical methods.
2- Any Troponin I concentration close or higher than 0.8 ng/mL for serum and plasma samples or 1.0 ng/mL for blood samples may suggest a possible AMI.
The time required for blood cTnI level to reach the upper limit of normal has been found to be 4-6 hours after the onset, and then remains elevated for 6-10 days in some cases. 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- **Use only fresh whole blood samples (< 4 hours) when test is performed with blood samples. Finger prick samples should be assayed just after collection.**
4- In case of high RF (rheumatoid factor) or CRP (C-reactive protein) concentrations (high levels indicate acute infections), the test could exceptionally show a positive result.
5- In case of delayed reading time, i.e. over 20-25 minutes, the test could also show sometimes 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- This format of test is to be only used with VEDALAB's rapid test readers.
8- If the reading time (20 minutes) is not strictly respected, wrong results will be obtained.
9- This format of test should not be used for visual reading.
10- 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.
11- Do not use the reader for measurements before at least 30 minutes warm-up after having switched on.

## X- BIBLIOGRAPHY

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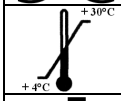
|---|---|---|---|
|  | Read the instructions before use |  | For *in vitro* diagnostic use |
|  | Temperature limitations |  | Do not reuse |
|  | Manufacturer | | |

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**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 change | - Chap VIII info c) sensitivity+ I) cross reaction (addition) + CI 95%+ g) intra assay reproducibility |
| Administrative | - Chap I (addition)<br>- Chap X bibliographic references 8& 9 (addition) |

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