# MGL-CHECK-1

## Ref. 15091

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

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

### I- PRINCIPLE

Myoglobin is an intra cellular heme protein involved in the storage and transfer of oxygen to muscle tissues (1). In man, myoglobin is one of the major proteins found in the myocardium and therefore it is not surprising that injury to cardiac muscle results in the release of myoglobin into the bloodstream (2, 3).

Normal levels in human serum are about 60-80 ng/mL with elevations as high as 1,500 ng/mL seen in patients with acute myocardial infarction (AMI). Consequently, myoglobin measurements are valuable indications in the early diagnosis of AMI as myoglobin has been reported to rise within 4 hours of the onset of symptoms (4, 5, 6). The MGL-CHECK-1 is a rapid quantitative assay for the detection of human myoglobin in serum, plasma or whole blood.
The method employs a unique combination of monoclonal-dye conjugate and polyclonal solid phase antibodies to identify myoglobin in the test samples with a high degree of specificity.

As the sample flows through the absorbent device, the labelled antibody-dye conjugate binds to the myoglobin forming an antibody antigen complex. This complex binds to the anti-myoglobin antibody in the reaction zone (T) and produces a pink-rose colour band. In the absence of myoglobin, there is no line in the reaction zone (T). The 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.

### II- MGL-CHECK-1 KIT COMPONENTS

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

| | | |
|---|---|---|
| 1- MGL-CHECK-1 reaction devices | 10 | 20 |
| 2- Disposable plastic pipettes | 10 | 20 |
| 3- Diluent in a dropper bottle | 2.5 mL | 5 mL |
| 4- Instructions leaflet | 1 | 1 |

**5- Controls (Optional):**

**Positive control (ref. V2600) and Negative control (ref. V2601)**: 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 and 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 MGL-CHECK-1 kit components should be stored at any temperature between +4°C to +30°C.
**2- <u>Do not freeze the test kit.</u>**
3- MGL-CHECK-1 is stable until the expiry stated on the package label.

### IV- 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.

### V- SPECIMEN COLLECTION AND PREPARATION

1- MGL-CHECK-1 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- <u>Whole blood samples should be tested immediately (< 4 hours). Finger prick samples should be assayed just after collection</u>.**
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.

### VI- ASSAY PROCEDURE

**a) Control testing**

- Wait for 15 minutes after freeze-dried dissolving.
- Add the requested volume (25µL) with **<u>lab pipette (disposable tips)</u>** 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 7 days after reconstitution**.

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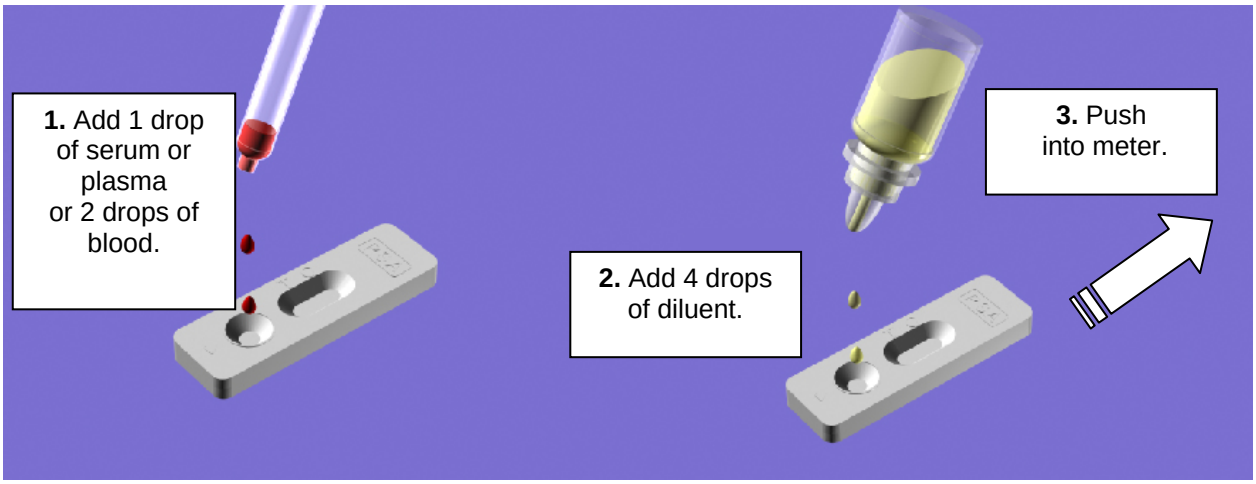
**b) Samples testing**

***<u>Follow the below instructions or refer to the picture n°1.</u>***

1– Allow samples and MGL-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 serum dropper with specimen (serum or plasma) and by holding it vertically, dispense one drop (25 µl) into sample well. (If whole blood is used, dispense 2 drops (50 µL) into the well (▷ ) **<u>and wait for the blood sample to be completely absorbed before adding diluent</u>**.
5– Add exactly 4 drops of diluent (150 µL) in the sample well (▷).
6– Read the result **(in ng/mL**) 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 tests readers, refer to the corresponding leaflet.




**Picture n°1**

## VII- PERFORMANCES CHARACTERISTICS

**a) Linearity**

The measuring range is 50-500 ng/mL.
For myoglobin concentration below 50 ng/mL, the result will be given as “< 50 ng/mL ”.
For myoglobin concentration over 500 ng/mL, the result will be given as “>500 ng/mL ”.
For samples whose concentration is higher than 500 ng/mL, dilute with saline and repeat the assay as per instructions of Part. VI.

**b) Sensitivity**

Concentrations close to 20 ng/mL are detected by MGL-CHECK-1 test. In these cases, results will be rendered as “<50 mg/mL”.
Levels higher than 80 ng/mL are generally considered as abnormal values.

**c) Accuracy**

A study has been performed using serum samples obtained from dilutions of commercially available Myoglobin antigen covering a range of 50 to 500 ng/mL. Optical densities expressed as a function of myoglobin concentrations are described by following polynomial curve :

$$Y = 30.57 + 1.71\ x - 2.06.10^{-3}\ x^2\ (r = 0.9284)$$

The results show a good correlation (r > 0.90) of the values obtained with MGL-CHECK-1 on VEDALAB’s reader.
A panel of 30 human sera samples pre-assayed on the ROCHE ELECSYS 2010 analyser have been tested with MGL-CHECK-1 rapid test. Results were read using VEDALAB’s reader. Results are shown in table I.

Table I

| Human sera identification Panel SCIPAC (lot: 565-146) | AGE | SEX | [MGL] in ng/mL Expected value (ROCHE ELECSYS 2010) | *Confidence range* | | [MGL] in ng/mL Obtained values (MGL-CHECK-1) |
|---|---|---|---|---|---|---|
| | | | | Lower limit | Upper Limit | |
| 44 | 51 | F | 47.3 | 35.5 | 59.1 | <50 |
| 41 | 51 | F | 78.09 | 58.6 | 97.6 | 71.2 |
| 47 | 51 | F | 82.82 | 62.1 | 103.5 | 64.6 |
| **31** | **83** | **M** | **95** | 71.2 | 118.7 | **<50** |
| 28 | 60 | M | 99.9 | 74.9 | 124.9 | 88.1 |
| 33 | 77 | M | 106.6 | 79.9 | 133.2 | 104.3 |
| 37 | 63 | F | 114.5 | 85.9 | 143.1 | 107.5 |
| 30 | 52 | F | 130.1 | 97.6 | 162.6 | 104.3 |
| 29 | 53 | F | 133.7 | 100.3 | 167.1 | 101.8 |
| 48 | 65 | F | 157.5 | 118.1 | 196.9 | 150.1 |
| 49 | 62 | M | 177.4 | 133 | 221.7 | 174.9 |
| 21 | 69 | F | 177.5 | 133.1 | 221.9 | 155.6 |
| 45 | 86 | F | 183.1 | 137.3 | 228.9 | 217.4 |
| 34 | 69 | M | 198.6 | 148.9 | 248.2 | 177.6 |
| 27 | 53 | F | 273.2 | 204.9 | 341.5 | 214.7 |
| 26 | 69 | F | 277 | 207.7 | 346.2 | 242.2 |
| 24 | 77 | M | 320.5 | 240.4 | 400.6 | 238.0 |
| **39** | **58** | **M** | **367.1** | 275.3 | 458.9 | **487.2** |
| 20 | 76 | M | 394.4 | 295.8 | 493 | 338.9 |
| 43 | 20 | F | 788.3 | 591.2 | 985.4 | >500 |
| 35 | 71 | F | 804.7 | 603.5 | 1005.9 | >500 |
| 42 | 17 | F | 867 | 650.2 | 1083.7 | >500 |
| 36 | 81 | F | 1050 | 787.5 | 1312.5 | >500 |
| 32 | 78 | F | 1477 | 1107.7 | 1846.2 | >500 |
| 40 | 74 | F | 2128 | 1596 | 2660 | >500 |
| 22 | 72 | M | >3000 | N/A | N/A | >500 |
| 23 | 49 | M | >3000 | N/A | N/A | >500 |
| 25 | 62 | M | >3000 | N/A | N/A | >500 |
| 38 | 72 | M | >3000 | N/A | N/A | >500 |
| 46 | 67 | F | >3000 | N/A | N/A | >500 |

N/A : Not Applicable.

A discrepancy is obtained with two sera samples (identified by bold typo):

-Serum 39 (strong positive): The results obtained by the both methods indicate the same clinical diagnosis profile (ie. Most probably myocardial infarction)

-Serum 31 (borderline): This sample has a high level of CRP revealing a recent infection and the probable presence of poly-specific antibodies that could interfere with immunodiagnostic reagents (MGL-CHECK-1 and ROCHE ELECSYS 2010).

Negative, borderline and positive samples are correctly detected (a correlation of 91.6% has been established between VEDALAB rapid test and ROCHE ELECSYS 2010).

**d) Intra-assay reproducibility**

Within run precision was evaluated by using 26 replicates of two commercially available references containing 65.7 and 203.8 ng/mL of myoglobin as determined with quantitative MGL-CHECK-1 for VEDALAB's readers.
The obtained CVs (coefficient of variation) were respectively equal to 9.52% and 9.64%.

## VIII- LIMITATIONS

1- As for any diagnostic procedure, the physician should confirm the data obtained using this test by other clinical methods.
2- The time required for blood myoglobin level to reach the upper limit of normal has been found to be 2-4 hours after the onset. Therefore, a negative result within the first hours of the onset of symptoms does not rule out AMI with certainly. 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- Some serum specimens with high rheumatoid factor concentration (RF) may yield non specific positive results during testing. Such cases should be discriminated before testing.
5- 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.
6- This format of test is to be only used with VEDALAB rapid test readers (EASY READER® or EASY READER+®).
7- If the reading time (15 minutes) is not strictly respected, wrong results will be obtained.
8- This format of test should not be used for visual reading.
9- 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.

## IX- BIBLIOGRAPHY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2- **Drexel, H et al.** Myoglobinaemia in the early diagnosis of acute myocardial infarction. Am Heart J 105 : 642-651, 1983.
3- **McComb, JM et al.** Myoglobin in the very early phase of acute myocardial infarction. Ann Clin. Biochem, 22 : 152-155, 1983.
4- **Grenadier E., Keidar S., Kahana L., Alpan G., Marmur A., Palant A :** The roles of serum myoglobin, total CPK, and CKMB isoenzyme in the acute phase of myocardial infarction. Am heart J. 1983. 105 : 408-416.
5- **Ellis AK, Saran BR,** 1982. Kinetics of myoglobin release and prediction of myocardial myoglobin depletion after coronary artery reperfusion. Circulation 1989. 80 : 676-683.
6- **McComb, JM et al**. Myoglobin and creatine Kinase in acute myocardial infarction. Br. Heart J, 1984. 51 : 189-194.


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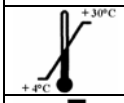
|---|---|---|---|
|  | Read the instructions before use |  | For *in vitro* diagnostic use |
|  | Temperature limitations |  | Do not reuse |
|  | Manufacturer | | |

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