C-Peptide-CHECK-1

Quantitative determination of C-Peptide in whole blood, plasma or serum samples

FOR EASY READER® OR EASY READER+® USE ONLY

Ref.: 160091 (20 tests) & Ref.:160091-10T (10 tests)

I- PRINCIPLE

Human C-Peptide, a 31 aminoacid residue peptide having a molecular mass of approximately 3 000Da. (1) connects Insulin's A and B chains within the proinsulin molecule (2, 3). Proinsulin is subsequently cleaved enzymatically into the beta-cells of the pancreas leading to an equimolar release of insulin and C-Peptide into the blood stream (4).

The degradation rate of C.Peptide in the body being slower than that of insulin (half life of 20-30 minutes, compared with the half-life of insulin of just 3-5 minutes), its measurement is a more stable indicator of insulin secretion and beta-cell function (5).

Due to its relative metabolic inertness, the C-Peptide levels in peripheral venous blood are about 5-6 times greater than insulin levels. In addition, another advantage of C-Peptide assay is its ability to distinguish endogenous from injected insulin. Therefore, a very low C-Peptide concentration confirms Type 1 diabetes and insulin dependence while elevated C-Peptide levels are associated to Type 2 diabetes or to the increased B-cell activity observed in insulinomas (6, 7, 8).

The C-Peptide-CHECK-1 test is a rapid quantitative assay for the detection of C-Peptide in serum, plasma or whole blood. The method employs a unique combination of monoclonal-gold conjugate and monoclonal solid phase antibodies to identify C-Peptide in the test samples with a high degree of specificity.

As the test sample flows through the absorbent device, the labelled antibody-gold conjugate binds to the C-Peptide forming an antibody-antigen complex. This complex binds to the anti- C-Peptide antibody in the positive reaction zone (T) and produces a pink-rose colour band.

In the absence of C-Peptide, there is no line in the positive reaction zone (T). The reaction mixture continues flowing through the absorbent device past the reaction zone (T) and control zone (C).

Depending on the C-Peptide concentration level, different lines of different intensities will appear on the reading window allowing the quantitative measurement of C-Peptide when used in combination with the VEDALAB's readers EASYREADER® or EASYREADER+®.

II- C-Peptide-CHECK-1 KIT COMPONENTS

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

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

III- STORAGE AND STABILITY

1- All C-Peptide-CHECK-1 kit components should be stored at room temperature (+4°C to +30°C) in the sealed pouch.

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

3- The C-Peptide-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 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- C-Peptide-CHECK-1 test is to be 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 negative specimen before testing.

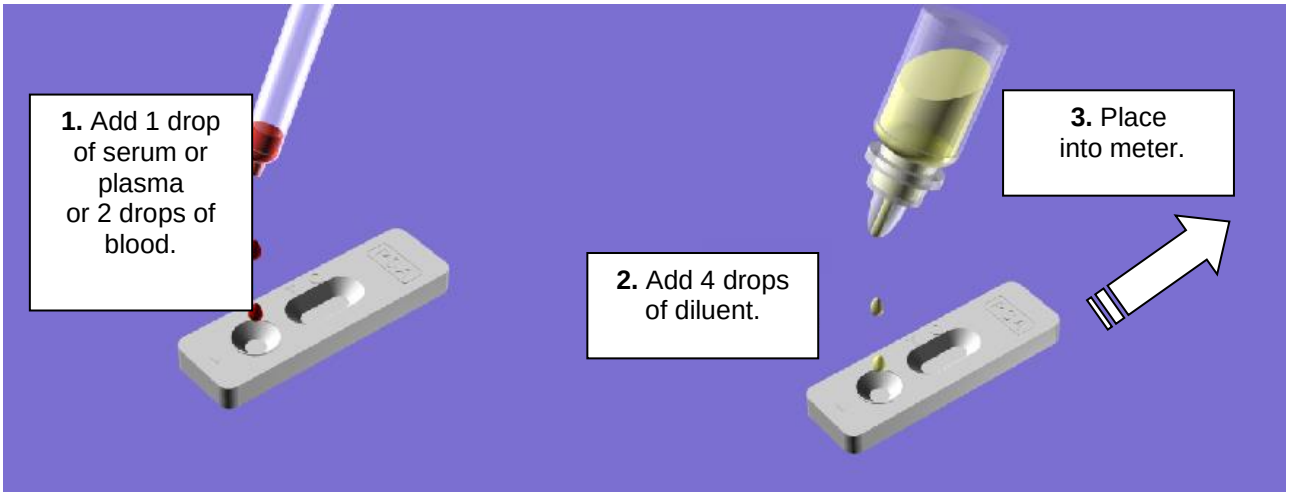
CE

VI- ASSAY PROCEDURE

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

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

1- Allow samples and C-Peptide-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 specimens (serum or plasma) and by holding it vertically, dispense one drop (25 µL) into sample well. If the whole blood is used, dispense two drops (50 µL) into the sample well (▷) **<u>and wait for the blood sample to be completely absorbed before adding diluent</u>**.
5- While keeping the diluent dropper bottle **<u>vertically</u>**, add slowly 4 drops of diluent (150 µL) in the sample well (▷) with an interval of 2-3 seconds in between each drop. Make sure that the tip of the dropper vial does not contact the sample well to avoid cross contamination.
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 corresponding leaflet.



Picture n°1

VII- PERFORMANCES CHARACTERISTICS

a) Linearity

The measuring range is 0.5-20ng/mL.
For C-Peptide concentration below 0.5ng/mL, the result will indicate "< 0.5ng/mL".
For C-Peptide concentration within a range of 20ng/mL to 40ng/mL, the result will be indicated as "20-40ng/mL".
For C-Peptide concentration over 40 ng/mL, the result will indicate "> 40 ng/mL".

b) Accuracy

A study has been performed using preassayed serum samples as well as C- peptide international standard containing C-Peptide concentration in a range of 0 to 40ng/mL. Optical density expressed as a function of C-Peptide concentration is described by following linear curve:

$$Y = 0.417x^2 + 29.59x + 18.36$$

The results show an excellent correlation ($r > 0.99$) of the values obtained with C-Peptide-CHECK-1 using VEDALAB's readers.

c) Analytical sensitivity

The analytical sensitivity of C-Peptide-CHECK-1 test is 0.5ng/mL.

d) Precision

A panel of 43 human serum samples preassayed using the ROCHE ELECSYS analyser was assayed using the C-PEPTIDE-CHECK-1 quantitative test. The obtained results show an overall correlation of 97.8%.

e) Expected values

It is recommended that each laboratory should determine its own normal and abnormal values. In normal healthy adults, a normal C-Peptide range is 1.1 to 4.4 ng/mL. The published literature indicates that reference intervals for adults and paediatric patients are comparable.

f) Intra assay reproducibility

Two pre assayed C-peptide samples (2ng/mL and 10ng/mL respectively) have been assayed (20 replicates each) and showed acceptable CV's (14% and 14.2% respectively).

g) Interferences

1- Bilirubin:

Low, medium and high C-peptide samples spiked with 0.03g/L of bilirubin showed repeatedly correct results.

2- Triglycerides:

Low, medium and high C-peptide samples spiked with 10g/L of triglycerides showed repeatedly correct results.

3-Hemoglobin:

Low, medium and high C-peptide samples spiked with 1.5g/L of hemoglobin showed repeatedly correct results.

h) Hook effect

Specimens containing very high levels of C-Peptide (up to 5,000ng/mL) when tested consistently showed positive results.

VIII- LIMITATIONS

1- As for any diagnostic procedure, the physician should evaluate data obtained by the use of this test in light of other clinical information.

2- Some serum specimens with high rheumatoid factor (RF) concentration may yield non-specific positive results during testing. Such cases should be identified before testing.

3- 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.

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

5- This format of test is to be only used with VEDALAB rapid test readers.

6- If the reading time (10 minutes) is not strictly respected, wrong results will be obtained.

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

8- 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.

9-Do not use the reader for measurements before at least 30 minutes warm-up after having switched on.

IX- BIBLIOGRAPHY

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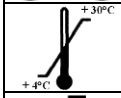
7. **Jones AG, Hattersley AT (Jul 2013).** "The clinical utility of C-Peptide measurement in the care of patients with diabetes". Diabetic Medicine. 30(7):803-17. doi:10.1111/dme. 12159. PMC3748788.
8. **Leighton E.**, **Sainsbury CAR, Jones GC**: A practical Review of C-Peptide testing in diabetes. Diabetes Ther 8:475, 2017.

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

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