ACE - liquid

UV enzymatic Method

| 4 x 25 ml | CL01-100 | |
|---|----------|--|
| Other available kits: ACE (lyophilized reagents) | CY02-36 | |
| Available for quality control: ACE-CONTROL SERUM N + P Control serums in normal and pathological range | 7508 | |
| ACE-CALIBRATOR For an accurate control of instrument calibration | 7512 | |
| ACE-STANDARD Standard of ACE for Measuring of the Enzyme in Serum | 7511 | |

INTENDED USE

Kit for the quantitative determination of angiotensin converting enzyme (ACE) in serum and plasma.

CLINICAL MEANING

The angiotensin-converting enzyme (ACE) is one of the main components of the renin-angiotensin system (RAS), which controls blood pressure by regulating the volume of bodily fluids. It also increases blood pressure through the constriction of blood vessels.

PRINCIPLE

The angiotensin converting enzyme (ACE) catalyzes hydrolysis of furylacryloylphenylalanylglycylglycine (FA-Phe-Ala-Gly-Gly) substrate to furylacryloyl phenyl-alanine and glycylglycine.

Hydrolysis is related to an absorbance decrease valued at 340 nm and is proportional to enzymatic activity.

SAMPLE

Serum or heparinized plasma.

ACE is a metalloprotein: it is mandatory to avoid using chelating agents (e.g EDTA) for the preparation of the sample.

STABILITY: 7 days at 2-8°C, 6 months at -20°C.

REAGENTS

| Package content | CL01-100 |
|--------------------|-----------|
| REAGENT 1 (lyo) | |
| FA-Phe-Ala-Gly-Gly | 4 x 25 ml |
| Buffer pH 8,4 | |

STABILITY: stored at 2-8°C and protected from light, reagents are stable up to the expiration date on the package.

NECESSARY ITEMS – NOT PROVIDED

Usual laboratory equipment: UV/VIS Spectrophotometer with temperature control; automatic micropipettes; Optical glass cuvettes or, alternatively, disposable ones in optical polystyrene; saline solution.

MANUAL ASSAY PROCEDURE

| Wave length: | 340 nm |
|-----------------------|-------------------------|
| Optical Path: | 1 cm |
| Reading: | Against distilled water |
| Temperature: | 37°C |
| Method: | Fixed time |
| Reaction time: | 15 minutes |
| Sample/Reagent Ratio: | 1/10 |

NOTE: spectrophotometric reading is made in a substrate spectrum zone where even a small wavelength change corresponds to a high variation of extinction coefficients.

For a proper use, carefully check the wavelength calibration and the instrument sensibility.

For this aim, use the product ACE CALIBRATOR.

Pipette into cuvette:

| Reagent 1 | 1.0 mL |
|-----------|--------|
| Sample | 0.1 mL |

Stir and incubate at 37°C. After 5 minutes read A1 absorbance and after exactly 15 minutes from the first reading, read A2 absorbance.

CALCULATION

ACE Activity (in U//L): = (A1-A2) x 863

NOTES

To calculate activity use the following formula: U/L = (A1 - A2) x [(Vt x 1000) / ($\Delta\epsilon$ x I x Vs X t)] where:

A1: absorbance in the sample after 5 minute incubation; A2: absorbance in the sample after 15 minute incubation from the first reading; Vt : total volume (reagent +sample) in ml; $\Delta \epsilon$: variation of extinction coefficient at 340 nm; I : optical path in cm; Vs : sample volume in ml; t : incubation time in minutes. Under these test conditions the formula becomes: U/L = (A1 - A2) x [(1,1 x 1000) / (0,85 x 1 x 0,1 x 15)] = = (A1 - A2) x 863 $\Delta \epsilon$ was defined by research spectrophotometers. Using chemical analysers, $\Delta \epsilon$ might reach a different value with following modification of U/L values in healthy and in ill people. Use ACE CALIBRATOR to calculate $\Delta \epsilon$ for the instrument used.

REFERENCE VALUES

| AVERAGE | ± | SD |
|----------|---|----------|
| 90 1 U/I | ± | 24.3 U/L |

Reaction volumes can be proportionally varied without any change in calculation. Each laboratory should define its own reference values for this method.

QUALITY CONTROL – CALIBRATION

All Clinical Chemistry laboratories should implement a quality control program. Control serums of human origin are available for this purpose on request: **ACE CONTROL SERUM N+P** with normal and pathological value ranges A Standard for an accurate control is also available: **ACE STANDARD 2x1 ml**

PERFORMANCE CHARACTERISTICS

Sensitivity

The method can discriminate up to 2.5 U/L

Linearity: up to 250 u/l

For higher values, dilute the sample 1:1 with saline solution, repeat the test and multiply the result by 2.

| Within run (n=10) | Average U/L | SD | CV% |
|--------------------|-------------|------|------|
| Sample 1 | 99 | 1.08 | 1.08 |
| Sample 2 | 191 | 1.76 | 0.92 |
| | 1 | | |
| Between run (n=20) | Average U/L | SD | CV% |
| Sample 1 | 99 | 2.33 | 2.35 |
| Sample 2 | 196 | 3 20 | 1.68 |

Correlation:

The kit shows a correlation coefficient equal to 0.989 in comparison to another kit available on the market.

Interferences

lt's not possible to verify interferences if: lipids < 900 mg/dl. Ascorbic acid <50 mg/dl

Avoid using chelating agents (EDTA) during the preparation of the sample.

DISPOSAL

The product must be used for professional analysis only. The product must be disposed of according to national/international laws.

WARNINGS AND PRECAUTIONS

The reagents may contain non-reactive components and various preservatives. Contact with the skin and ingestion should be avoided. Use the normal precautions expected with correct behaviour in laboratory.

REFERENCES

1. Harjanne A. Clin. Chem. 30 (1984) 901

MANUFACTURER

FAR

Via Fermi, 12 - 37026 Pescantina - VERONA - ITALY tel +39 045 6700870 website <u>http://www.fardiag.com</u> o.mail: order@fardiag.com

e-mail: <u>order@fardiag.com</u> e-mail: <u>fardiag@fardiag.com</u>

KEY SYMBOLS

| IVD | In Vitro diagnostic medical device |
|-------------|------------------------------------|
| LOT | batch number |
| REF | catalogue number |
| X | temperature limits |
| Σ | use by |
| \triangle | caution |
| Ĩ | consult accompanying documents |

Issue 01 - Jan 2021 RR