



ACE (Angiotensin Converting Enzyme)

KINETIC COLORIMETRIC DETERMINATION IN SERUM AND PLASMA
 FAPGG (N-[3-(2-furyl)acryloyl]-L-phenylalanyl-glycylglycine) METHOD
 For in vitro diagnostic use only

Kit: 10 x 10 ml

Cod. ACE 8865

SUMMARY

ACE is an hydrolase that transforms Angiotensin I (quite inactive) in Angiotensin II (very strong vasoconstrictor).
 ACE is also able to inactivate bradykinin.
 Elevated levels of ACE occur in patients with active sarcoidosis, with tuberculosis, Gaucher's disease and in many other pathological conditions in lung and liver diseases.

PRINCIPLE

The ACE present in the serum catalyzes the hydrolysis of the FAPGG, forming furylacryloylphenylalanine (FAP).
 The decrease of absorbance in the unit time at 340 nm is proportional at the activity of the ACE in the sample.

REAGENT

Component of the kit: **Cod. ACE 8865**
***REAGENT 1** (liquid) **2 x 50 ml**
 Good's Buffer >20 mmol/L pH 8.2
***REAGENT 2** (lyophilized) **10 x 10 ml**
 FAPGG > 0.25 mmol/L

STABILITY: the reagents, stored at 2-8°C, are stable up to the expiry date shown on the package **if not contaminated during handling.**

AUXILIARY REAGENTS FOR CALIBRATION CURVE

In order to have a Calibration Curve, we suggest the use of the following kit:

- **ACE CALIBRATOR 6 x 1 mL**
Cod. ACAL8866 (see the related insert)

AUXILIARY REAGENTS FOR QUALITY CONTROL

The reliability of test results should be monitored by routine use of artificial urine Controls of known concentrations. We suggest following kits:

- **ACE NORMAL CONTROL kit 6 x 1 mL**
Cod. ANOR8867 (see the related insert)

- **ACE ELEVATED CONTROL kit 6 x 1 mL**
Cod. AEL8868 (see the related insert)

PREPARATION OF THE WORKING REAGENT

***KIT 10 x 10 ml (Cod. ACE 8865)**
 Add 10 ml of *Reagent 1 to one vial of *Reagent 2.
 Mix gently until dissolution.

STABILITY: the reconstituted *Reagent 2 stored at 2-8°C, is stable up to 4 weeks **if not contaminated during handling.**

Close immediately after handling.
Incompetent handling will release us from any responsibility.

SAMPLE

• Not haemolyzed serum, plasma with heparin. EDTA cannot be used.

PROCEDURE

• Wavelength: 340 nm
 • Pathlength: 1 cm
 • Reading: against air or distilled water
 • Temperature: 37°C
 • Method: kinetic
 • Reaction: 5 + 5 minutes
 • Linearity: up to 150 U/l a 37°C
 • Sample/Reagent: 1/10

Let reagent reaches the working temperature before using.

Pipette in test tube or cuvette so labelled:
 ST: Standard/Calibrator; S: Sample:

	S	ST
Working Reag.	1000 µl	1000 µl
Sample	100 µl	----
Standard	----	100 µl

Mix wells, incubate for 5 minutes at 37°C.
 Read the absorbance of standard (Ast1) and sample (As1).
 Exactly after 5 minutes at 37°C read again standard (Ast2) and sample (As2).

Determine the diff. of absorbance for sample and calibrator:

$$\Delta As = A1s - A2s$$

$$\Delta Ast = A1st - A2st$$

CALCULATION

$(\Delta As / \Delta Ast) \times \text{Calibrator conc.} = \text{U/L of ACE.}$

REFERENCE VALUES

	37°C	30°C
U/L	8 - 52	5 - 33

It is suitable that every laboratory determines its normal reference values.

PERFORMANCE CHARACTERISTICS

These performance characteristics was determined using a spectrophotometer or analyzers typically found in clinical laboratories, under the stated assay conditions.

Linearity: ACE may be determined between 3 - 150 U/L.
 For concentrations ≥ 150 U/L, dilute the sample 1:4 with saline sol., repeat the determination and multiply the result $\times 4$.

Sensitivity: The minimum detectable is 3 U/L.

Within-run Precision:

	Mean (U/L) $\pm 2s$	CV %
Serum 1	50,2 \pm 4,8	4,77
Serum 2	119,8 \pm 6,9	2,86

Run-to-run (Day-to-day) Precision:

	Mean (U/L) $\pm 2s$	CV %
Serum 1	49,9 \pm 6,9	6,21
Serum 2	120,0 \pm 8,4	3,51

Interferences: See References point 2.

Correlation: A group of 20 sera from 3 to 130 U/L was assayed by this procedure and using a similar commercially available ACE Reagent.
 Comparison of the data gave following results:

Linear regression equation $y = 1,0563x - 0,8$
 Correlation coefficient $r = 0,9988$

NOTE

1. A proportional. variation of the reaction volumes does not change the result.
2. We suggest do not mix Reagents from different Production lots.
3. Dilute the sample with activity higher than 150 U/L, with saline solution 1:4; repeat the determination and multiply the result by 4.

4. Very deep attention must be given to interfering substances: certain drugs and other substances are able to influence levels of ACE (see References 2.).

5. PAY ATTENTION!

Applications on routine Analyzers may be totally different from what we developed as manual determination, and also from themselves.

6. The reagent must be used only for the intended destinations, by expert people and in the due lab. conditions.

7. The clinical diagnosis cannot be done using the result of only one test, but have to be done integrating different lab. and clinical data.

8. Avoid the use of anticoagulants containing fluorides and EDTA.

REFERENCES

1. Textbook of Clinical Chemistry, Ed. by N.W. Tietz, W.B. Saunders Co., Philadelphia (1999).
2. Young D.S. et al., Clin. Chem. 21, 302D (1975).
3. Maguire G.A. et al., Ann. Clin. Biochem. 22, 204 (1985).

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