ACE. Direct. UV Ref.:ACE-101L
Determination of Angiotensin Converting Enzyme in serum and plasma
Only for *in vitro* use in clinical laboratory
Store at 2-8ºC 20 mL

ACE

CE

PRINCIPLE OF THE METHOD
ACE reagent is for use in the determination of angiotensin converting enzyme (ACE) activity in serum or plasma at 340 nm.
The following reaction is catalysed by ACE:
FAPGG → FAP + Glycylglycine

FAPGG (furylacryloylphenylalanine-glycylglycine) is hydrolysed to furylacryloylphenylalanine (FAP) and glycylglycine. Hydrolysis of FAPGG results in a decrease in absorbance at 340 nm. The ACE activity in the sample is determined by comparing the samplereaction rate to that obtained with the ACE calibrator.

CLINICAL SIGNIFICANCE
Angiotensin converting enzyme (ACE, dipeptidylcarboxypeptidase) is a glycoprotein peptidyldipeptide hydrolase that cleaves histidylleucine dipeptide from angiotensin I, a relatively inactive decapeptide. The latter is converted to the potent vasoconstrictor, angiotensin II. ACE also inactivates bradykinin. Elevated levels of ACE activity occur in serum of patients with active sarcoidosis, and occasionally in premature infants with respiratory distress syndrome, in adults with tuberculosis, Gaucher's disease, leprosy, and in many other pathologic conditions involving lung and liver diseases.

REAGENTS

R 1	Goods buffer pH 8.2	80 mmol/L
	FAPGG	0.5 mmol/L,
	Sodium azide	< 0.1 %

PREPARATION
- **R 1** is ready to use.

STORAGE AND STABILITY
All the components of the kit are stable until the expiration date on the label when stored tightly closed at 2-8ºC and contaminations are prevented during their use. Do not freeze the reagents.
- **R 1**: on board 30days if contamination is avoided.

Signs of reagent deterioration:
- Presence of particles and turbidity.

ADDITIONAL EQUIPMENT
- Spectrophotometer or colorimeter measuring at 340 nm.
- Matched cuvettes 1.0 cm light path.
- General laboratory equipment.

SAMPLES
Fresh serum or plasma (lithium- or sodium-heparin) promptly separated from the red blood cells.
Stability of the sample: 7 days at 2-8 °C or **1** year at -20 °C.

Do not use:
1) lipemic samples
2) hemolyzed samples
3) EDTA as an anticoagulant as it inhibits the ACE activity

PROCEDURE
1. Assay conditions:
   Wavelength: . . . . Main = 340 nm / Secondary= 660 nm
   Cuvette: . . . . 1 cm light path
   Temperature 37ºC
2. Adjust the instrument to zero with distilled water.
3. Pipette into a cuvette:

	Calibrator	Control	Sample
R 1 (µL)	1000	1000	1000
Calibrator (µL)	100	-	--
Control (µl)	-	100	-
Sample (µL)		--	100

4.Mix and incubate for 4 min at 37ºC.
5. Read the absorbances 1.
6. Incubate for 5 min. at 37ºC.
7. Read the absorbances 2.

CALCULATIONS
$\frac{\text{ODsample - ODblank}}{\text{ODcalibrator - ODblank}}$ x Calibrator value (U/L)

Conversion factor:
ACE (U/L) x 0.001 = ACE (kU/L)
ACE (U/L) x 0.01667 = ACE (µkat/L)

QUALITY CONTROL
Control sera are recommended to monitor the performance of assay procedures.
If control values are found outside the defined range, check the instrument, reagents and calibrator for problems.
Each laboratory should establish its own Quality Control scheme and corrective actions if controls do not meet the acceptable tolerances.

REFERENCE VALUES[2]

8 – 65 U/L

These values are for orientation purpose; each laboratory should establish its own reference range.

PERFORMANCE CHARACTERISTICS
Measuring range: 5.4 – 160 U/L .
Samples with concentration higher than 160 U/L must be diluted 1:10 with distilled water and result multiplied by 10.

Precision:

	Intra-assay		Inter-assay	
Mean (U/L)	26.3	88.9	27.0	94.0
SD	0.9	1.79	1.01	3.80
CV (%)	3.42	2.01	3.71	3.97

Sensitivity: 2.4 U/L.

Accuracy: Results obtained using our reagents (y) did not show systematic differences when compared with other commercial reagents (x).
The results obtained using 143 samples were the following:
Correlation coefficient (r): 0.976.
Regression equation: y= 0.98x – 0.56
The results of the performance characteristics depend on the analyzer used.

INTERFERENCES
The test is not affected by the presence of unconjugated bilirubin up to **13** mg/dL, conjugated bilirubin up to **26** mg/dL, haemoglobin up to **100** mg/dL and triglycerides up to **200** mg/dL.
Captopril, an ACE inhibitory drug, used for the treatment of hypertension and some types of congestive heart failure, will inhibit ACE activity in serum or plasma11.

NOTES
We have instruction sheets for several automatic analyzers. Instructions for many of them are available on request.

BIBLIOGRAPHY

1) NCCLS Procedures for the Collection of Diagnostic Blood Specimens by Venipuncture; Approved Standard – Fifth Edition (H3-A5). Wayne, PA: The National Committee for Clinical Laboratory Standards, 2003.
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3) US Department of Health and Human Services. Biosafety in Microbiological and Biomedical Laboratories, 5th ed. Washington,DC: US Government Printing Office, January 2007.
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5) Sewell DL, Bove KE, Callihan DR, et al. Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline - Third Edition (M29-A3). Wayne, PA: Clinical and Laboratory Standards Institute, 2005.
6) Pesce, A.J., Kaplan, L.A.: "Methods in Clinical Chemistry", Mosby Ed. (1987).
7) Burtis C.A., Ashwood E.R.: "Tietz Textbook of Clinical Chemistry", W.B. Saunders Company Ed. (3rd edition, 1999).