Quantitative determination of Homocysteine in serum or plasma
Only for *In Vitro* Diagnostic use

ORDER INFORMATION

REF	Cont.
HCY 20	1 X 20 ML
HCY 40	1 X 40 ML

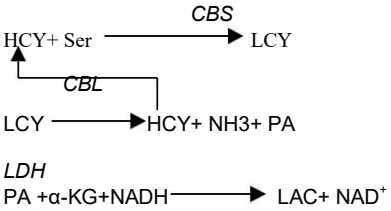
CLINICAL SIGNIFICANCE

Homocysteine as an independent cardiovascular risk index has been widely accepted, is another risk factor for high cholesterol, smoking, diabetes outside. Homocysteine is a sulfur-containing amino acid produced by the metabolism of methionine. Hcy levels and cardiovascular disease are closely related. Hcy increased blood vessel wall because of irritation caused by damage to the arteries, eventually causing heart blood flow is blocked. High homocystinuria patients because of severe genetic defects affect Hcy metabolism, resulting in high Hcy hyperlipidemia. Slight genetic defects or nutritional deficiencies of B vitamins will be accompanied by moderate or mild elevated Hcy also increase the risk of heart disease. Hcy increased distortion and can cause neural tube birth defects such as congenital malformation diseases.

Method

Photometric Enzymatic Test method

PRINCIPLE



REAGENT

Reagent I : Buffer Reagent
Reagent II : Enzyme Reagent
Calibrator : refer vial label

REAGENT PREPARATION

Use separate reagents ready to use.

REAGENT STORAGE AND STABILITY

Unopened, avoid light preservation in 2 ~ 8 ℃, valid for 12 months; Opened, avoid light preservation in 2 ~ 8 ℃, valid for 1 month.
Reagent is not allowed frozen.

WARNING AND PRECAUTIONS

- For in vitro diagnostic use.
- Do not use components beyond the expiration date.
- Do not mix materials from different kit lot numbers.
- Exercise the normal precautions required for handling all laboratory reagents.
- The reagent contains preservative. Do not swallow. Avoid contact with skin and mucous membranes.
- For detailed information refer Material Safety Data Sheet.

WASTE MANAGEMENT

Please refer to local legal requirements.

MATERIALS REQUIRED BUT NOT PROVIDED

- NaCl solution 9 g/L
- General laboratory equipment

SAMPLE COLLECTION AND PRESERVATION

Serum or Heparin anticoagulant Plasma
It is best to fresh serum or heparin anticoagulant blood plasma, once take, blood immediately centrifugal separation of plasma.
It is very important to store the sample protected from light!
Stability:
7 days at 4 – 8°C
Don't use the blood sample collection bottle containing sodium fluoride. Before take blood, please try to avoid high protein diet, high protein diet may lead to increase Homocysteine

ASSAY PROCEDURE

Operating Instructions

- Check reagent inventories at least daily to ensure that quantities are sufficient for the planned work load.
- Bring all reagents, standard and samples to room temperature 18 - 28 ºC, prior to analysis.

AUTOMATED PARAMETERS	
Primary Wavelength	340nm
Secondary Wavelength	405nm
Cuvette Light Path	1 cm
Reaction Type	Fixed Time
Reaction Temperature	37°C
Reaction Direction	Decreasing
Sample Volume	13 µl
Reagent 1 Volume	240 µl
Reagent 2 Volume	65 µl
Delay Time	90 Seconds
Read Time	180 Seconds
Linearity	0 – 50 µmol/L

MULTI POINT CALIBRATION

Multi Point calibrator Available on request (Optional)
Prepare the following HCY calibrator dilutions in NaCl 9 g/dL, Multiply the concentration of the HCY calibrator by the corresponding factor stated in the table below to obtain the HCY concentration of each dilution.

Calibrator Dilution	1	2	3	4	5
Calibrator HCY (µL)	-	25	50	75	100
NaCl 9 g/dL (µL)	100	75	50	25	-
Factor	0	0.25	0.50	0.75	1.0

MANUAL ASSAY PROCEDURE

Addition Sequence	Calibr (C)	Test (T)
Reagent 1	240 µL	240 µL
Calibrator	13 µL	-
Sample	-	13 µL
Mix well and incubate for 5 minutes at 37° C		
Reagent 2	65 µL	65 µL

Mix well, and read the absorbance after 90 sec A1 and after 180 sec minutes A2 of the sample/calibrator addition.

SAMPLE DILUTIONS

- This method is linear upto a concentration of 50 µmol/L.
- Dilute samples above this concentration 1:1 with 0.9% saline
- Repeat assay. Multiply the result by 2.

CALCULATION

Sample Concentration = $\frac{\text{Sample } \Delta\text{Abs/min}}{\text{Calibrator } \Delta\text{Abs/min}} \times \text{Calibrator Concentration}$

CALIBRATORS AND CONTROLS

For the calibration of automated photometric systems the commercially available suitable multi-calibrator is recommended.

It is recommended to run a normal and a pathological control serum which is commercially available to verify the performance of the

measured procedure. The value of controls should fall within the established limit.

Each laboratory should establish corrective action in case of deviations in control recovery.

PERFORMANCE CHARACTERISTICS

WITHIN RUN

Sample	Mean Concentration	SD	CV %
Norm	8.18	0.19	2.34%
Path	28.33	0.74	2.60%

RUN TO RUN

Sample	Mean Concentration	SD	CV %
Norm	8.36	0.15	1.76%
Path	28.83	0.78	2.69

LINEARITY

The method is linear upto a concentration of 50 µmol/L. Dilute samples above this concentration 1:1 with 0.9% saline solution and repeat assay. Multiply the result by 2.

Limit of detection: The limit of detection for Homocysteine is 0.02 µmol/L.

METHOD COMPARISON

A comparison of Accucare Bilirubin Total with a commercially available assay (x) using 20 samples gave following results: $R^2 = 0.9900$

REFERENCE VALUES

In plasma, Homocysteine age-related in the normal reference range,
adult≤15µmol/L,
Above 60 years 15~20µmol/L,
Above 100 years 25~27µmol/L.

The reference values are to be considered as indicative only. Every laboratory should establish its own normal range.

LIMITATION OF THE PROCEDURE

- HCY testing is just one of the standard that clinicist diagnose the patient. Clinical physicians should according to patients' bodies, history and other diagnostic program, to get comprehensive judgment.
- For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

INTERFERENCE

- In sample, blood ammonia ≤50µmol/L; glutathione ≤0.5mmol/L; The elf sulfide concentration ≤20µmol/; adenosine ≤100µmol/L; bilirubin ≤20 mg/dL、hemoglobin ≤1200 mg/dL; triglyceride ≤2500mg/dL; ascorbic acid≤10mmol/L; S-adenosylmethionine（SAM）≤20µmol/L; L- cysteine≤1.0mmol/L, not observed clearly obvious interference.
- These characteristics have been obtained using an automatic analyzer. Results may vary if a different instrument or a manual procedure is used.

BIBLIOGRAPHY

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GLOSSARY OF SYMBOL

	Consult Instruction for Use	LOT	Lot Number
REF	Catalog Number		Date of Manufacturing
	Store between		Use By or Expiration Date
	Manufacturer	IVD	For *in vitro* Diagnostic use only
	Keep away from sunlight	CONT	Content of the kit

LAB-CARE DIAGNOSTICS (INDIA) PVT. LTD.
C1 Type, Shed No.: 3225, Chemical Zone,
GIDC Sarigam – 396155, Dist. Valsad, Gujarat, India.
Tel.: +91 22 2554 2109 /1558
Email: accucarediagnostics.com; Website: www.labcarediagnostics.com