Pariksha Biotech
A game changer in IVD

hS CRP Estimation Kit

High-Q hsCRP

Latex Enhanced Turbidimetric Immuno Assay (LETIA)

HIGH®
Command on Quality

Intended Use:
The high sensitivity C-Reactive Protein (hsCRP) assay is for the in vitro quantitative determination of C-Reactive protein (CRP) in human serum and plasma on automated clinical chemistry analyzers. Measurement of CRP is of use for the detection and evaluation of inflammatory disorders and associated diseases, infection and tissue injury.

Clinical Significance:
CRP (C-reactive protein, MW=25106Da) is an acute phase protein whose concentration is seen to increase as a result of the inflammatory process, most notably in response to pneumococcal (bacterial) infectious, histolytic disease, and a variety of disease conditions. Originally discovered by Tillet et al. in 1930 in patient sera with acute infection, CRP has now to be used as a marker or general diagnostic indicator of infections and inflammation, in addition to serving as a monitor of patient response to therapy and surgery. Furthermore, regular measurements of CRP in infants can be a useful aid in the early diagnosis of infectious diseases.

Principle of the test:
The hsCRP assay is based on a Latex Enhanced Turbidimetric Immuno Assay (LETIA). When an antigen-antibody reaction occurs between CRP in a sample and Anti-CRP which has been sensitized to latex particles, agglutination results. This agglutination is detected as an absorbance change (546 nm) with the magnitude of the change being proportional to the quantity of CRP in the sample. The actual concentration is then determined by the interpolation from a calibration curve prepared from calibrators of known concentration.

Kit Composition
R-1:
100 mM Tris-buffer solution
0.09% Sodium Azide
R-2:
Suspension of latex particles (0.5%) coated with goat anti human CRP, 0.09% Sodium Azide

Warnings and Precaustions:
Store the reagents at 2-8°C. DO NOT FREEZE. Avoid contact with skin and eyes. Contains sodium azide, which may react with lead or copper plumbing to form explosive compounds. Flush drains with copious amounts of water when disposing of this reagent. Specimens containing human sourced materials should be handled as if potentially infectious, using safe laboratory procedures such as those outlined in Biosafety in Microbiological and Biomedical Laboratories. Additional safety information concerning storage and handling of this product is provided within the Material Safety Data Sheet for this product.

Reagent Preparation
All the reagents are ready to use and do not need any preparation. Swirl the Latex Reagent gently before use .
CRP Calibrators: Liquid Stable and do not need any reconstitution

Reagent stability and storage:
The hsCRP assay reagents should be stored at 2-8°C. DO NOT FREEZE. The reagents are stable when stored as instructed until the expiration date on the label. Do not mix reagent components from different lots.

Specimen collection and preparation:
Serum or heparinized plasma or EDTA plasma samples can be used for the hsCRP assay.
For serum, collect whole blood by venipuncture and allow clotting.
For plasma, mix the sample by gentle inversion prior to centrifugation. Centrifuge and separate serum or plasma as soon as possible after collection. Sample stability 6 to 11 days at room temperature (15-25°C); 2 months at 2-8°C; and 3 years at -20°C It is recommended that frozen samples are thawed at room temperature; samples must be mixed well before analysis. Repeated freezing and thawing should be avoided.

Kit Configuration:
Materials Supplied

Reagent 1	1 x 40 mL
Reagent 2	1 x 10 mL
CRP Calibrators	4 x 0.5 ml

Quality Control:
We recommend that each laboratory should use CRP controls to validate the performance of hsCRP reagents. A set of HIGH-Q hS-CRP controls is available separately. The range of acceptable control limits should be established by individual laboratories.

Results:
Results are printed out in mg/L.
Note: Samples with values greater than 150.0 mg/dl should be diluted with saline and rerun. Multiply results by the dilution factor.

Accuracy / Correlation:
Correlation studies were performed by testing 57 serum samples with CRP concentrations ranging from 0.2 to 185mg/L in comparison with an existing commercial CRP assay method. The linear regression gives a correlation r2 value of 0.990, slope of 1.01, and y intercept of 0.0196.

LOB, LOD, and LOQ:
The LOB, LOD, LOQ of the hsCRP Assay was determined on the Hitachi 917 according to CLSI EP17-A. By testing a True Blank Sample (7.5% BSA) in 20 replicates daily for 3 days, LOB was determined to be 0.08 mg/L. By testing five low serum samples (100 x diluted) in 4 replicates for 3 days, LOD was determined to be 0.13 mg/L. To determine LOQ, specimens with mean measured concentrations ranging from 0.118 to 0.978 mg/L were assayed. Based on the EP evaluator- 8 fitted model, the LOQ (lowest concentration for which CV is less than a target of 20% with 95% of confidence interval) is 0.20 mg/L CRP.

Assay Range	0.10 – 150 mg/L
Lower Limit of Detection	0.13 mg/L
Functional Sensitivity	To at least 0.15 mg/L

PROCEDURE:
Pipette into test tubes labeled Calibrator (C) and Test (T).

(Reagent)	(Calibrator)	(Test Sample)
Reagent-1	400 µl	400 µl
CRP Calibrators(1,2,3,4)	5 µl	-
Sample	-	5 µl
Reagent-2	100 µl	100 µl

Reaction temperature : 37°C

Mix well and read absorbances of Calibrators (C) and Test (T) against distilled water at 546 nm (530-550 nm) as follows:

Initial absorbance A1 -exactly after 5 sec.
Final absorbance A2 - exactly 240 sec. after A1determine Δ A for Calibrators (C) and Test (T)

CALCULATIONS :

$$\text{CRP Conc.: (mg/L)} = \frac{\text{(A2-A1) Sample}}{\text{(A2-A1) Calibrators}} \times \text{Calibrators Concentrations}$$

System Parameters:

Reaction Type	:	Fixed Time / Multi Standard / Linear / Spline
Reaction Direction	:	Increasing
Sample Volume	:	5 µl
Reagent Volume	:	Reagent-1 400 µl+ Reagent-2 100 µl
Wave Length	:	546nm (530-550 nm)
Calibrators Conc.	:	On the labels
Flow Cell Temp.	:	37°C
Linearity	:	150
Zero setting with	:	Distilled Water
Units	:	mg/L
Delay	:	5 sec.
Measuring	:	240 sec

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Linearity:
CRP linearity set was prepared by diluting a specimen containing 400 mg/dl CRP with saline according to CLSI EP6-A. Assay linearity was tested on the Hitachi 917. Data analysis using EP Evaluator 8 showed that the hsCRP assay was linear through a measured range of 0.10 to 150 mg/L with an allowable systematic error of 4.5%.

Expected Values:
The assay reference interval was determined using serum specimens from 103 apparently healthy adults with ages of 18- 62 according to CLSI C28-A3 guideline. The serum specimens were tested in duplicate by the hsCRP method. EP Evaluator 8 Software was used to verify the reference interval.
C-Reactive Protein is a non-specific indicator for a wide range of disease processes.
Reference intervals may be affected by different factors.

Expected values:

Recommended cardiac risk assessment categories:

Low Risk for CVD	**< 1 mg/L**
Intermediate Risk for CVD	**1.0 to 3.0 mg/L**
High Risk for CVD	**> 3.0 mg/L**

Newborns with no evidence of infection have CRP concentrations < 1 mg/L

General Reference Values to rule out active infection and general inflammatory response <6 mg/L

Risk Stratification:
For patients with acute coronary syndromes, measurement of hsCRP may provide prognostic information. A value of CRP > 10 mg/L in the early period (6 - 24 hours after onset of symptoms), has been shown to be indicative of an increased risk for short term (30 days - 1 year) recurrent cardiac events substudy of 447 patients in the CAPTURE trial examined the clinical implications of elevated levels of CRP for risk stratification in patients with unstable angina. As shown in the following graph, patients with a CRP >10 mg/L experienced a higher event rate (mortality or MI) than patients with a CRP <10 mg/L

Because of the variation depending on age, sex, diet, and geographical location, each laboratory should determine its own expected values for the different patient groups as dictated by good laboratory practice.

AHA/CDC Expert Panel Recommendations :
hsCRP levels should not be substituted for assessment of traditional cardiovascular risk factors. Application of management guidelines for acute coronary syndromes should not be dependent on hsCRP levels.In patients with stable coronary disease or acute coronary syndromes, hsCRP measurement may be useful as an independent marker of prognosis.When using the assay for risk assessment, patients with persistently unexplained, marked elevation of hsCRP (>10 mg/L) after repeated testing should be evaluated for non-cardiovascular etiologies. The expert panel recommends against screening of the entire adult population for hsCRP as a public health measure. Patients with evidence of active infection, systemic inflammatory processes or trauma should not be tested for cardiovascular disease risk assessment until these conditions have abated. Application of secondary prevention measures should not depend on hsCRP determination, but rather an array of risk factors (global risk assessment). Serial measurements of CRP should not be used to monitor effects of treatment. Two separate CRP measurements (optimally two weeks apart) should be obtained before performing risk assessment, due to within-subject CRP variability. Measurement of hsCRP is an independent marker of risk. hsCRP levels may be useful in motivating patients to improve lifestyle behaviors.

References:

1. Knidmark C-O: The concentration of C-reactive protein in sera from healthy individuals, Scand J. Clin Lab Invest 29: 407-411, 1972.
2. Du Clos TW: Function of C-reactive protein, Ann Med 32:274-278, 2000.
3. Tietz, N.W. (Ed.), Clinical Guide to Laboratory Tests, 4th Edition, Alan H. Wu, Saunders Elsevier (2006).
4. Maksimowicz-McKinnon K, Bhatt DL, and Calabrese LH:Recent advances in vascular inflammation: C-reactive protein and other inflammatory biomarkers, Curr. Opin.Rheumatol 16: 18-24, 2004.
5. Pearson TA, Mensah GA, Alexander RW, et al. Markers of inflammation and cardiovascular diseases; application to clinical and public health practice: A statement for healthcare professionals from the Centers for Disease Control and Prevention and the American Heart Association. Circulation 2003; 107: 499-511.
6. Use of anticoagulants in Diagnostic laboratory Investigations.WHO Publication WHO/DIL/LAB 99.1/Rev. 2 Jan. 2002.
7. Benitz, W.E., et al., Serial serum C-reactive protein levels in the diagnosis of neonatal infection. Pediatrics 1998;102:E41

Ordering information

Cat No:	Pack Size	Presentation
P-hsCRP-50	50 ml	(40.0 ml R1 + 10 ml R2 with 4 Calibrators)

Product Features

- Two liquid reagents
- 4 Level calibrator set provided
- Linearity : 150 mg/L
- Multipoint Linear Assay
- 4 Minutes Assay (5 Sec Delay and 240 Sec Measuring)
- No Prozone effect was detected upon 100 mg/L
- Bilirrubin (20 mg/dl), lipemia (10 g/l) and rheumatoid factors (300 IU/ml) do not interfere. Hemoglobin (≥ 5 g/l), interferes.
- Can be used on semi and fully auto analyzers

Symbols used with IVD devices

	Date of manufacture		Manufactured by
IVD	In vitro diagnostic device		Keep away from sunlight
	Do not freeze		This way up
	Use by (yyyy-mm-dd or mm/yyyy)	R	Reagent
CAL	Calibrator Material	LOT	Batch code
2°C 8°C	Temperature limitation (store at)	CONTROL	Control
	Consult instructions for use		Keep dry Keep away from rain
REF	Catalog Number		

eIFU Indicator

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SCAN TO EXPLORE MORE

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