KAISER PERMANENTE®
COLORADO LABORATORY

C-Reactive Protein - VITROS 350

PRINCIPLE:

The VITROS CRP Slide is a multilayered, analytical element coated on a polyester support.

The immuno-rate format for CRP is based on an enzymatic heterogeneous, sandwich immunoassay format. In this format a derivative of phosphorylcholine (PC) is covalently bound to polystyrene polymer beads and in the presence of calcium serves as a capture agent. Monoclonal anti-CRP antibody conjugated to horseradish peroxidase (HRP) serves as a signal generator.

A drop of patient sample is deposited on the slide and is evenly distributed by the spreading layer to the underlying layers. CRP in the sample binds to PC-linked capture beads and anti-CRP antibody labeled with horseradish peroxidase to form an insoluble sandwich complex in Incubation 1. The subsequent addition of 12 µL of VITROS Immuno-Wash Fluid to the slide removes unbound materials from the read area, while also providing the hydrogen peroxide required for the enzyme-mediated oxidation of leuco dye. The reflection density of the dye is measured after the addition of VITROS Immuno-Wash Fluid at the end of Incubation 2. This reflection density is directly proportional to the concentration of CRP in the sample. To determine if an adequate wash has occurred, the wash detection dye is read at 540 nm immediately after Incubation 2..For further details, see individual method sheet in the VITROS "Instructions for Use Manual".

`

SCOPE:

Medical Technologists and Medical Laboratory Technicians

SPECIMEN REQUIREMENTS:

1 Serum, Heparinized plasma or EDTA
2 Specimens are retained in stoppered containers for 5 days in the specimen refrigerator. Specimens are stable for 3 days at 2-8 degrees C; room temperature 18-28 degrees C for 4 hours or less; frozen at -18 degrees C for six months.

EQUIPMENT AND MATERIALS:

- VITROS CRP Slides
- VITROS Calibrator Kit 7
- Quality Control Materials, 3 Levels (see Quality Control Material Procedure)
- VITROS Chemistry Products Specialty Diluent
- VITROS Chemistry Products Immuno-Wash Fluid
- VITROS Chemistry Products FS Diluent Pack 3 (Specialty Diluent/Water) (for on-analyzer dilution)
- Sample cups

Written: J. Scherer 8/13
Revised:

REAGENTS:

1. **Slide Ingredients:** Immobilized phosphorylcholine 0.07 mg; mouse anti-CRP antibody labeled with horseradish peroxidase 0.0006 U; calcium chloride 0.08 mg; and 2-(3,5-dimethoxy-4-hydroxyphenyl)-4,5-bis(4-dimethylaminophenyl) imidazole (leuco dye) 0.04 mg.
2. ***Inactive ingredients:*** Binders, buffer, surfactants, cross-linking agent, polymer beads, proteins, stabilizers and wash detection dye.

Cartridge Handling, Storage and Stability:

1. **Warm at room temperature for 60 minutes in the foil wrapper before putting on the instrument.**
2. **Store frozen at -18 degrees C or below.**
3. **Stable on instrument for 48 hours.**
4. CRP Slides are stable until the expiration date on the carton when they are stored and handled as specified.
5. **Note: Do not use slide cartridges with damaged or incompletely sealed packaging.** Inspect the packaging for damage and use caution when opening to avoid damaging package.
6. Unwrap and load the cartridge into the Vitros slide supply.

CALIBRATION:

1 VITROS Chemistry Calibration Kit 7.
2 Store unopened – frozen at -18degrees C or below.
3 Store reconstituted at 2-8 degrees C. Stable for 24 hr.

See Calibration section of VITROS Operations Manual.

QUALITY CONTROL:

For Quality Control Procedure and Materials used, Reporting Ranges and other operational details, see that section of the Vitros procedure manual.

PROCEDURE:

Instrument Operating Instructions:

Refer to the operating instructions for the VITROS 350 Chemistry System.

****IMPORTANT:**

Bring all fluids and samples to room temperature, 18°.28°C, prior to analysis.

Written: J. Scherer 8/13
Revised:

Sample Dilution

Serum and Plasma

If CRP concentration exceeds the system's measurement range: *

Manual Sample Dilution

1. Dilute the sample with VITROS Specialty Diluent or a patient sample containing a low concentration of CRP. An initial threefold dilution is recommended.
2. Reanalyze.
3. Multiply the results by the dilution factor to obtain an estimate of the original sample's C-reactive protein concentration.

On-Analyzer dilution

Not recommended

REPORTING RESULTS:

Results are transmitted to the LIS and if necessary, may be reported directly from the Vitros printout.

REFERENCE RANGE:

Adults 0 to 1.00 mg/dL

LIMITATIONS OF THE PROCEDURE:

Interfering substances:

•Specimens with low total protein <4.9 g/dL (49 g/L) may give a positive bias of greater than +0.31 mg/dL (+3.1 mg/L) at a CRP concentration of 2.0 mg/dL (20.0 mg/L).

•Specimens with an elevated total protein >9.5 g/dL (95 g/L) may give a negative bias greater than -0.31 mg/dL (-3.1 mg/L) at a CRP concentration of 2.0 mg/dL (20.0 mg/L).

Interferent *	Interferent			CRP Concentration		Bias	
	Concentration		Comments	Conv. (mg/dL)	SI (mg/L)	Conv. (mg/dL)	SI (mg/L)
Ampicillin	200 mg/dL	(5.7 mmol/L)	Therapeutic	2.4	24	-0.52	-5.2
Gentisic acid	5 mg/dL	(0.32 mmol/L)	Therapeutic	2.5	25	-0.54	-5.4
Hemoglobin	400 mg/dL	(4 g/L)		2.0	20	+0.24	+2.4

DISTRIBUTION:

1 KP Laboratory Website - Policies and Procedure - MOL Chemistry Section
2 Regional Reference Laboratory - QA Manager Document Control

REFERENCES:

1.Tietz NW (ed). *Fundamentals of Clinical Chemistry*. ed. 3. Philadelphia: WB Saunders; 328.329; 1987.
2. Tietz NW. *Textbook of Clinical Chemistry*. Philadelphia: WB Saunders; 589; 1986.
3. Young DS. *Effects of Drugs on Clinical Laboratory Tests*. ed. 4. Washington D.C.: AACC
4. VITROS CRP Slides Test Methodology sheet.

Written: J. Scherer 8/13
Revised: