COD 31321 1 x 20 mL	COD 31921 1 x 50 mL	COD 31029 2 x 200 mL
STORE AT 2-8ºC		
Reagents for measurement of CRP concentration Only for *in vitro* use in the clinical laboratory		

C-REACTIVE PROTEIN (CRP)

BioSystems REAGENTS & INSTRUMENTS

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C-REACTIVE PROTEIN (CRP)
LATEX

PRINCIPLE OF THE METHOD

Serum C-reactive protein (CRP) causes agglutination of the latex particles coated with anti-human C-reactive protein. The agglutination of the latex particles is proportional to the CRP concentration and can be measured by turbidimetry[1-4].

CONTENTS

	COD 31321	COD 31921	COD 31029
A. Reagent	1 x 16 mL	1 x 40 mL	2 x 160 mL
B. Reagent	1 x 4 mL	1 x 10 mL	2 x 40 mL

COMPOSITION

A. Reagent: Glycine buffer 0.1 mol/L, sodium azide 0.95 g/L, pH 8.6.

B. Reagent: Suspension of latex particles coated with anti-human CRP antibodies, sodium azide 0.95 g/L.

STORAGE

Store at 2-8ºC.

Reagents are stable until the expiry date shown on the label when stored tightly closed and if contaminations are prevented during their use.

Indications of deterioration:

– Reagents: absorbance of the blank over 0.900 at 540 nm.

AUXILIARY REAGENTS

S. CRP Standard: 1 x 1 mL (BioSystems Cod. 31113). Human serum. C-reactive protein concentration is stated on the vial label. The concentration value of the CRP Standard is traceable to the ERM-DA472/IFCC (Institute for Reference Materials and Measurements, IRMM).
Human serum used in the preparation of the standard has been tested and found to be negative for the presence of antibodies anti-HIV and anti-HCV, as well as for HBs antigen. However, the standard should be handled cautiously as potentially infectious.

Reconstitute with 1.0 mL of distilled water. Stable for 1 month at 2-8ºC.

REAGENT PREPARATION

Working Reagent: Pour the contents of a Reagent B vial into a Reagent A bottle (Note 1). Mix thoroughly. Stable for 60 days at 2-8ºC.

Smaller Working Reagent volumes can be prepared by mixing: 1 mL of Reagent B + 4 mL of Reagent A. Shake the Reagent B vial before pipetting.

ADDITIONAL EQUIPMENT

– Thermostatic water bath at 37ºC

– Analyzer, spectrophotometer or photometer thermostatable at 37ºC able to read at 540 ± 20 nm.

SAMPLES

Serum collected by standard procedures.

CRP in serum is stable for 7 days at 2-8ºC.

PROCEDURE

1. Bring the Working Reagent and the instrument to 37ºC.
2. Zero the instrument with distilled water (Note 2)
3. Pipette into a cuvette:

Working Reagent	1.0 mL
Standard (S) or Sample	7 µL

4. Mix and immediately insert cuvette into the instrument. Start stopwatch.
5. Record the absorbance at 540 nm after 10 seconds (A_1) and after 2 minutes (A_2).

CALIBRATION

A calibration is recommended at least every 2 months, after reagent lot change or as required by quality control procedures.

CALCULATIONS

The CRP concentration in the sample is calculated using the following general formula:

$$\frac{(A_2\text{-}A_1)_{\text{Sample}}}{(A_2\text{-}A_1)_{\text{Standard}}} \times C_{\text{Standard}} = C_{\text{Sample}}\ (\text{mg/L})$$

REFERENCE VALUES

Serum, adults[5]: Up to 5 mg/L.

This range is given for orientation only; each laboratory should establish its own reference range.

QUALITY CONTROL

It is recommended to use the Rheumatoid Control Serum level I (Cod. 31213) and II (Cod. 31214) to verify the performance of the measurement procedure.

Each laboratory should establish its own internal Quality Control scheme and procedures for corrective action if controls do not recover within the acceptable tolerances.

METROLOGICAL CHARACTERISTICS

– Detection limit: 1.0 mg/L.

– Linearity limit: 150 mg/L. For higher values dilute sample 1/5 with distilled water and repeat measurement. Linearity limit may vary depending on the photometer or analyzer used. (Note 3)

– Repeatibility (within run):

Mean concentration	CV	n
7.4 mg/L	4.5 %	20
19.0 mg/L	3.6 %	20

– Reproducibility (run to run):

Mean concentration	CV	n
7.4 mg/L	4.6 %	25
19.0 mg/L	3.7 %	25

– Trueness: Results obtained with this reagent did not show systematic differences when compared with reference reagents. Details of the comparison experiments are available on request.

– Zone effect: This method has not zone effect (< 500 mg/L).

– Interferences: Hemoglobin (10 g/L), bilirubin (20 mg/dL), lipemia (triglycerides 10 g/L) and rheumatoid factors (200 IU/mL) do not interfere. Other drugs and substances may interfere[6].

These metrological characteristics have been obtained using an analyzer. Results may vary if a different instrument or a manual procedure are used.

DIAGNOSTIC CHARACTERISTICS

C-Reactive Protein (CRP), which is synthesized in the liver, is one of the the most sensitive acute phase reactants after tissue damage or inflammation. CRP activates the classical complement pathway as a response to the inflammatory reaction.

CRP levels in plasma can rise dramatically after myocardial infarction, stress, trauma, infection, inflammation, surgery or neoplastic proliferation. The increase occurs within 24 to 48 hours and the level may be 2000 times normal. An elevation can be expected in virtually all diseases involving tissue damages so the finding is nonspecific[7].

Clinical diagnosis should not be made on the findings of a single test result, but should integrate both clinical and laboratory data.

NOTES

1. Shake the Reagent B vial gently before pouring its contents into the Reagent A bottle. It is advisable to wash the Reagent B vial with a small volume of the prepared mixture in order to completely rinse the vial and avoid any losses.
2. These reagents may be used in several automatic analysers. Instructions for many of them are available on request.
3. The linearity limit depends on the sample to reagent ratio. It will be higher by decreasing the sample volume, although the sensitivity of the test will be proportionally decreased.

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