



# Canine C-Reactive Protein (cCRP) BIO-ANALYSER

Product Number: RL0002

## Assay Precautions

- Do not use kit or individual reagents past their expiry date.
- Do not mix or substitute reagents from different kit batches.
- Samples should be stored refrigerated or frozen if they are not to be analysed shortly after collection.
- Avoid repeat freeze/thawing of samples.
- Where possible avoid the use of haemolysed or lipaemic serum.
- Cover or cap all reagents when not in use unless stored on board analyser.
- Use clean and preferably disposable labware for all reagent preparation.
- Care must be taken not to contaminate reagents. Use fresh tips for each sample and reagent.

## Safety Precautions

- For *in vitro* research purposes only.
- Dispose of all clinical samples, infected or potentially infectious material in accordance with good laboratory practice.
- Wear disposable gloves and safety glasses where appropriate.
- The kit contains reagents that may cause irritation to skin and eye. Any reagent which comes into contact with the skin or eye should be washed off with water immediately.
- The kit contains reagents that may cause irritation to respiratory or gastrointestinal tract if inhaled or ingested. Seek medical attention if you feel unwell.



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## Intended Use

The canine C-Reactive Protein (cCRP) turbidimetric assay is intended for the quantitative measurement of CRP in canine serum.

## Clinical Use

Acute phase proteins (APPs) such as canine CRP are serum proteins that increase in concentration 24 to 48 hours following infection, inflammation, or trauma. The circulating concentrations of these proteins can provide an objective measure of the health status of an animal and are increasingly being used as markers of animal health and welfare.

Serum concentrations of APPs are related to the severity of the underlying condition, and provide a ready means of evaluating both the presence and extent of disease.

In a healthy animal, CRP is absent from serum or present in very low levels ranging from less than or equal to 10 mg/L in dogs.

## Methodology

The cCRP bio-analyser assay is an immunoturbidimetric assay. CRP in a sample of canine serum is captured by Reagent 1 and 2 to form an antibody-antigen complex.

The mixture is then measured turbidimetrically by the spectrophotometer in the biochemical analyser. The absorbance is directly proportional to the amount of cCRP present in the serum sample.

By comparison to standards with known concentrations of CRP the assay is calibrated. It is recommended that a calibration is performed once every 4 weeks, with quality controls (QCs) run once a day.

## Reagents Provided

- Reagent 1 (R1)
  - 1 x 60 mL
- Reagent 2 (R2)
  - 1 x 20 mL
- cCRP calibration set
  - 6 x 0.6 mL
- cCRP QC set
  - 3 x 2.4 mL

## Additional Materials Required

- Biochemical analyser, preferably an Olympus AU series.
- Sample cups and racks.
- A variety of micropipettes and disposable tips capable of dispensing 10 µL – 1000 µL.
- Distilled water.

## Sample Preparation

The serum should be separated from the red blood cells as soon as possible after collection. Samples should be frozen if analysis cannot be performed immediately.

Samples will be diluted 1 in 25 in H<sub>2</sub>O (on board analyser using a pre-dilution programme – see Analyser Parameters section) before analysis.

Samples should be run neat and if required, a further dilution of 1 in 4 is recommended (40uL Sample + 120 uL d.H<sub>2</sub>O).



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## Storage and Stability

The contents of the canine CRP bio-analyser kit should be stored at 2-8°C and used within the expiry date detailed on packaging.

Individual components are labelled with individual expiry dates; however the kit should be used within expiry date detailed on packaging.

## Reagent Preparation

- R1 and R2 reagents are provided ready to use.

For use, open (a record can be made of the open date) and place bottles directly onto the biochemical analyser.

- Calibrators set is provided at the required concentrations; please see individual bottle labels for calibrator concentrations.

For use, remove from storage, dispense required volume into sample cup and return to storage.

- QC set contains a Low, Mid and High QC.

For use, remove from storage, dispense required volume into sample cup and return to storage.

## Procedure

- Place R1 reagent and R2 reagent onto the biochemical analyser.
- Dispense the required volume of calibrator and QCs.
- Perform calibration and QCs;
  - If QCs are within range dispense sample into sample cup, add to analyser and analyse.
  - If QCs are outside range re-run the QCs.
  - If QCs remain outside range re-calibrate and re-run QCs.

## QC Acceptance Ranges

Please see label associated with each QC for batch specific ranges.

NB – The stated ranges are applicable for Olympus AU series biochemical analysers. For any other analyser data, please contact us.

## Performance Characteristics

Analyser	Correlation (R <sup>2</sup> )	Within run imprecision (%)	Limit of detection (mg/L)
Pentra 400	0.9	<4	0.2
Prestige 24i	1.0	<2	0.1
Olympus AU series (1:25 pre-dilution programme)	1.0	<2	6 (0.2 at 1:25 dilution)

Analyser	Interference Levels
Pentra 400	No interference from Haemoglobin up to 1g/L, Triglyceride up to 0.5g/L and Bilirubin up to 50uM.
Prestige 24i	No interference from Haemoglobin up to 1g/L, Triglyceride up to 0.5g/L and Bilirubin up to 50uM.
Olympus AU series (1:25 predilution programme)	No interference from Haemoglobin up to 1g/L, Triglyceride up to 0.5g/L and Bilirubin up to 50uM.



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## Analyser Parameters Olympus AU Series

## Repeat Parameters

### General

	Volume (µl)		Pre-dilution Rate
Sample	5	with 10µl H2O	25
R1	135	-	-
R2	40	with 10µl H2O	-

Primary Wavelength	Secondary Wavelength
600nm	0nm

Method	Reaction Slope
End	+

Measuring point 1		Measuring point 2	
First	Last	First	Last
12	27	-	-

Reagent OD Limit			
First L	Last L	First H	Last H
-2	-2	2.5	2.5

Dynamic Range	
L	H
0*	X#

\*if repeat parameters required set to 7

#Refer to Calibrator 6 concentration (batch dependant)

Correlation Factor		On board stability period	
A	B		
1	0	30	

CALIBRATION		
Cal Type	Measure Type	Formula
6AB	Rack	POLYGONAL

Diluted - not required

	Volume (µl)		Pre-dilution Rate
Sample			

Condensed

	Volume (µl)		Pre-dilution Rate
Sample	5	with 10µl diluent	5

Normal

	Volume (µl)		Pre-dilution Rate
Sample	5	with 10µl diluent	25

Repeat Decision Level	L:-99999.99
	H: 99999.99