CALCIUM CPC LR liquid reagent

REF E1806100 6x100 ml
E1800650 6x50 ml
E1800450 4x50 ml

CE IVD For in vitro medical device

Use

Calcium Test is intended for the quantitative in vitro diagnostic determination of calcium in human serum, plasma and urine. Colorimetric o-cresolphtalein complexone method CPC.

Summary

Calcium measurementes are used in the diagnosis and treatment of certain disorders of calcium metabolism.

Principle

End point analysis. Under alcaline condictions O-cresolphtalein complexone (CPC) reacts with calcium ions and magnesium to form a purple complex :the magnesium interference is inhibited by 8-hydroxyquinoline presence. Absorbance measurements are taken at 570 nm. The increase in absorbance due to purple complex is directly proportional to the calcium concentration in the tested sample.

Reagents

R1	AMP buffer pH 10.3	30.0 mmol/l
R2	O-cresolphtalein complexone	0.16 mmol/l
	8-hydroxyquinoline	9.0 mmol/l

Reagent Preparation

Reagents are liquid and ready to use. About using as monoreagent ("sample-starter" procedure) mix the reagents **R1** and **R2** in equal parts.

Storage and stability

- Store the kit at 15-25°C.
- After opening, the vials R1, R2 are stable 90 days if recapped immediately and protected from contamination, evaporation, direct light, and stored at the correct temperature.
- Working solution stability (**R1**+ **R2**): 1 day at 15-25°C.

Precaution in Use

The product is not classified as dangerous (DLg. N. 285 art. 28 l. n. 128/1998). The final concentration of the components is below the limits imposed by Regulation (EC) No. 1272/2008 - CLP (and subsequent amendments) and Directive 88/379/CEE and subsequent amendments to the classification-packaging and labeling of dangerous substances.
However the reagent should be handled with caution, according to good laboratory practice.

Waste Management

Please refer to the local legal requirements.

Specimen Collection and Preparation

- Serum or plasma.
- Urine 24h Diluted: 1:5.
- Do not use samples with haemolysis.
- Do not use anticoagulants as EDTA, oxalate, citrate or fluoride.
- Avoid venous standstill. The use of tourniquet can rise the calcium level in the withdrawal in 0.5 mg/dl (0.12 mmol/l) as well.
- After picked up, serum and plasma must be separated, as soon as possible, from red cells to avoid calcium absorption by red cells.
- The calcium is stable in the serum or plasma 1 day at 2-8°C or 6 months at -20°C.

Note

- The kit, according to this method, must be used in manual procedures. About automatic using follow specific applications.
- Avoid direct light, contamination and evaporation.
- The volumes in the procedure can be changed proportionally.
- In case of complaint or quality control request, refer to the lot number on the package or the lot number on the singles vials.

Procedure

Wavelength	λ: 570 (550-590) nm
Working Temperature	37°C°
Optical path	1 cm
Reaction	"end point" (increasing)

- Monoreagent Procedure "sample starter"

	BLANK	STD	SAMPLE
Working Reagent	1000 µl	1000 µl	1000 µl
Distilled Water	25 µl	--	--
Sample	--	--	25 µl
Standard	--	25 µl	--

Mix, then incubate for 5' at 37°C. Measure the absorbance of the sample (EC) and standard (ESTD) against the reagent blank.

- Bireagent Procedure "substrate starter"

	BLANK	STD	SAMPLE
Reagent R1	500 µl	500 µl	500 µl
Distilled Water	25 µl	--	--
Sample	--	--	25 µl
Standard	--	25 µl	--

Mix, incubate at 15-25°C for 1' and then add:

Reagent R2	500 µl	500 µl	500 µl

Mix, then incubate for 5' at 37°C. Measure the absorbance of the sample (EC) and standard (ESTD) against the reagent blank.

Calculation

Calcium [mg/dl] o [mmol/l] = EC/ESTD x Conc. STD

Dluted urines: multiply the result for diluting factor.
The reagent performances are related to 37°C, 1 cm and 570 nm.

Conversion Factor

Calcium [mg/dl] x 0.2495 = Calcium [mmol/l]
Calcium [mg/dl] x 0.4990 = Calcium [mEq/l]

Reference Values

Adults	8.5 -10.8 mg/dl (2.11 – 2.68 mmol/l)
Children	8.5 – 12 mg/dl (2.11 – 2.98 mmol/l)
Urine	100 - 300 mg/24 h

Reference values are considered indicative since each laboratory should establish reference ranges for its own patient population. The analytical results should be evaluated with other information coming from patient's clinical history

ANALYTICAL PERFORMANCES

The performance of the reagent are related to 37 ° C, 1 cm and 570 nm

Linearity

The reaction is linear in concentration range between 0.56 e 15 mg/dl (0.138 - 3.74 mmol/l) . Samples with values exceeding 15 mg/dl must be diluted with saline solution. Then, multiply the result for the diluting factor.

"Intra-Assay" precision (within-Run)

Determined on 20 samples for each control(N-H) (Normal- High). Results:

MEAN (mg/dl)	N = 8.90	H= 14.40
S.D.	N = 0.12	H = 0.25
C.V.%	N = 1.38	H = 1.73

"Inter-Assay" precision (between-Run)

Determined on 20 samples for each control(N-H) (Normal- High). Results:

MEAN (mg/dl)	N =8.94	H= 14.08
S.D.	N =0.19	H = 0.26
C.V.%	N =2.11	H = 1.81

Analytical sensitivity

The test sensitivity in terms of detection limit is: 0.56 mg/dl (0.138 mmol/l).

Correlation

A study based comparing this method with a similar method on 20 samples has given a correlating factor **r = 0.962**

y = 0.933x + 0.8075

Interferences

No interference was observed by the presence of:

Bilirubin	≤ 35 mg/dl
Triglycerides	≤ 1500 mg/dl
Hemoglobin	≤ 200 mg/dl
Magnesium	≤ 15 mg/dl
Ascorbate acid	≤ 25 mg/dl

Quality controls

It's necessary, each time the kit is used, to make the quality controls and to check that values obtained are within the acceptance range provided in the insert.Each laboratory should establish its own mean and standard deviation and adopt a quality control program to monitor laboratory testing.

Bibliography

Ray Sarkar, B.C. et al., Anal. Bioch., 20,155, (1967).
Barnett, R.N. et al., Amer. J. Clin. Oath., 59, 836, (1973).
Kaplan, L.A., Pesce, A..J.: "Clinical Chemistry", Mosby Ed. (1996).

Symbols

CE	CE Mark (98/79 CE regulation)
IVD	in vitro medical device
LOT	Batch Code
	Use by
	Storage temperature limits
	Read instruction for use
	Gesan Production srl