Calcium

OCPC Method

Code : 10005 (2 x 60 ml)

(For the analyser/Colorimetric estimation of Calcium in Serum or Plasma)
In VITRO USE Only.

SUMMARY & EXPLANATION OF TEST:

The metabolism of Calcium is closely related with the metabolism of phosphorus. More than 99% of the Calcium in the body is present in bones as Calcium Phosphate. The reminder of Calcium, although a very small amount, is present in plasma and has varied and significant functions in the body. Calcium in plasma occurs in two forms : as non - diffusible protein bound Calcium and as free or ionized. Serum Calcium levels are also dependent upon the protein concentration.

For many years the estimation of Calcium was done by precipitation as Oxalate followed by titration. However, this method is time consuming and requires a large quantity of serum. Many colorimetric methods have now been developed. O-Cresolphthalein Complexone (OCPC) method was introduced by Anderegg, which was later modified by several workers to improve it's overall performance.

Calcium Kit is based on the method of Baginski in which serum is directly treated with O-Cresolphthalein Complexone reagent. The reagents are specially stabilized. The method is rapid, simple, and ideal for routine use in clinical laboratory.

PRINCIPLE:

Calcium in alkaline medium reacts with O-Cresolphthalein Complexone to form a purple colored complex whose absorbance is proportional to the Calcium concentration. Interference due to magnesium and iron is eliminated by using 8-hydroxyquinoline.

REAGENTS:

1. Buffer Solution 60 ml.
2. Colour Reagent 60 ml.
3. Standard (10 mg%) 3 ml.

The reagents are stable at 2 - 8°C till the expire date mentioned on the label.

SAMPLE :

Serum or Heparinized Plasma.Serum Calcium is stable for 24 hours at R.T. and for one week at 2 - 8°C.

EXPECTED RANGE:

Serum Calcium: 8.7 - 11.0 mg%

LINEARITY:

This method is linear upto 20 mg%. Samples exceeding 20 mg% should be dilute and reassayed. The result has to be multiplied by the dilution factor.

INSTRUCTIONS:

1. Contamination of glassware with Calcium, usually from detergents, is a problem in this assay. Therefore, glassware should be washed with dilute hydrochloric acid (30% V/V) and rinsed with high purity deionized water before use. This includes cleanliness of the cuvettes.
2. Plasma collected in EDTA, Citrate or Oxalate as an anticoagulant should not be used.
3. Serum should be seperated from the clot without delay.
4. For serum, fasting blood should be collected since lipemic or turbid serum could lead to erroneous results.

DIRECTIONS FOR USE ON ANALYSERS:

Reaction Type	:	End point with std
Reaction Slope	:	Increasing
Wave Length	:	570 nm (Yellow filter)
Incubation Temp.	:	Room Temperature
Incubation Time	:	5 mins
Standard	:	10 mg%
Linearity	:	20 mg%
Unit	:	mg%

PROCEDURE:

Pipette into clean dry test tubes labelled Blank (B) Standard (S) and Test (T)

	B	S	T
Buffer Solution(l)	2.0ml	2.0ml	2.0ml
Color Reagent(2)	2.0ml	2.0ml	2.0ml
Deionized water	0.02ml	...	...
Standard (3)	...	0.02rnl	...
Serum	...	...	0.02ml

Mix well and incubate at room temperature for five minutes. Measure the absorbance of Standard(S) and Test (T) against Blank (B) either on a photocolorimeter with Yellow filter or on Spectrophotometer at 570 nm, within 30 minutes.

CALCULATIONS:

$$\text{Calcium in mg\%} = \frac{\text{A of (T)}}{\text{A of (S)}} \text{ X 10 (Std. Conc)}$$

SI conversion factor
mMol/L = mg% X 0.25

NOTES:

★ Due to variations in inter - laboratory assay conditions, instruments and demography, it is recommended that each laboratory should establish its own normal range. To ensure adequate quality control, each run should include a normal and abnormal assayed controls. The assigned value of the control must be confirmed by this methodology.

★ Final diagnosis should be based on a co-relation of test results with other clinical observations / Diagnostic tools.

BIBLIOGRAPHY:

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2. Carl A. Burtis & Edward R. Ashwood "Mineral and Bone Metabolism" in Tietz Fundamentals of Clinical Chemistry, W.B. Saunders and company, Philadelphia, PA (1996) 685-703.
3. Gitelman, H.J., (1967), Anal Biochem 18:521.
4. Baginski, E.S., (1973), Clin Chem. Acta 46 : 46.

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