

Calcium Determination

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Introduction:

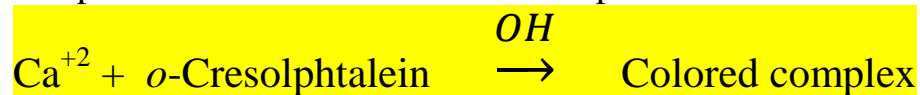
Calcium is required for cell function overall and for bone metabolism. Too little calcium gets you either a loss of tissue function or soft bones (osteoporosis).

While too much gives you tetanus (cardiac arrest and/or lock jaw is from over clenching of muscles). Changes in calcium are used to assess bone function.

Higher blood levels usually mean lower bone levels. Usually performed in conjunction with Phosphorous determinations.

PRINCIPLE OF THE METHOD

The measurement of calcium in the sample is based on formation of color complex between calcium and *o*-cresolphthalein in alkaline medium:



O-Cresolphthalein Complex one gives violet color in alkaline medium. The intensity of the colour formed is proportional to the calcium concentration in the Sample.

PREPARATION

All the reagents are ready to use. To prepare monoreagent, mix according to this proportion: 50 vol. of R1 and 1 vol. of R2.

SAMPLES -

Serum or plasma: Separated from cells as rapidly as possible. Blood anticoagulants with oxalate or EDTA are not acceptable since these chemicals will strongly chelate calcium.

- Urine: Collect 24 hour urine specimen in calcium free containers. The collecting bottles should contain 10 ml of diluted Nitric acid (50% v/v). Record the volume.

Dilute a sample 1/2 in distilled water. Mix. Multiply results by 2 (dilution factor). Stability of the samples: Calcium is stable 10 days at 2-8°C.

PROCEDURE

1. Assay conditions:

Wavelength: 570 nm (550-590)

Cuvette:..... 1 cm. light path

Temperature 37°C / 15-25°C

2. Adjust the instrument to zero with distilled water

3. Pipette into a cuvette:

	Blank	Standard	Sample
R1 (mL)	1.0	1.0	1.0
Standard (μL)	-----	10	-----
Sample (μL)	-----	-----	10

4. Mix and incubate for 5 min at 37°C / 15-25°C.

5. Read the absorbance (A) of the samples and calibrator, against the Blank. The color is stable for at least 40 minutes.

CALCULATIONS

Serum and plasma (A) Sample / (A) Standard x 10 (Standard conc.) = mg/dL calcium

Conversion factor: mg/dL x 0.25 = mmol/L.

REFERENCE VALUES

Serum or plasma:

Adults 8.5-10.5 mg /dL

Children 10 -12

❖ Hypocalcemia

- Hypoparathyroidism,
- Vitamin D deficiency
- Not enough calcium or vitamin D in your diet
- Irregular magnesium or phosphate levels
- Kidney disease
- Diabetes in the mother, in the case of infants

❖ **Hypercalcemia**

• Your body uses the interaction between calcium, vitamin D, and parathyroid hormone (PTH) to regulate calcium levels:

- Hyperparathyroidism
- Lung diseases and cancers
- Medication side effects

Some medications, particularly diuretics, can produce hypercalcemia

- Dehydration