

SAFETY PRECAUTIONS AND WARNINGS:

This reagent is for *In vitro* diagnostic use only.

INTENDED USE:

This reagent kit is intended for "*in vitro*" quantita

CLINICAL SIGNIFICANCE:

In the human body 98-99% of calcium is present in bound form in bones and teeth. About 50% of the blood calcium circulates in ionic form, the other part as bound to proteins. The Concentration of ionic calcium is influenced by the acid-base household of the body. The ratio of ionic/protein-bound calcium is higher in acidosis and lower in alkalosis. Elevated calcium levels are found in association with primary hyperparathyroidism, neoplastic diseases (eg. breast cancer, bronchial cancer, pancreatic tumor), osteoporosis, Paget's disease and Addison's disease, overdosage of the vitamins A and D, hyperthyroidism. Lower calcium values are measured in hypoparathyroidism, disturbances of the absorption, chronic renal failure, nephrotic syndrome, hepatic cirrhosis, acute pancreatitis.

PRINCIPLE:

At a neutral pH, the Ca²⁺ forms with arsenazo III a complex, the color intensity of which is directly proportional to the concentration Of calcium in the sample.

REAGENT COMPOSITION:

Reagent 1: Arsenazo III reagent Calcium
Standard: 10 mg/dl

MATERIALS REQUIRED BUT NOT PROVIDED:

- Clean & Dry Glassware.
- Micropipettes & Tips.
- Colorimeter or Bio-Chemistry Analyzer.

SAMPLES:

Serum free of hemolysis.
Urine diluted in ratio of 1:3 with distilled water, adjust to pH 3-4 with 0.1N HCl.

STABILITY OF REAGENT:

When stored tightly closed at room temperature protected from Light and contaminations prevented dur during their use; reagents are stable up to the expiry date stated on the label.

WORKING REAGENT:

The Reagent is ready for use.

GENERAL SYSTEM PARAMETERS:

| | |
|------------------------|------------------------|
| REACTION TYPE | End Point (Increasing) |
| WAVE LENGTH | 650 nm |
| LIGHT PATH | 1 cm |
| REACTION TEMPERATURE | 37°C |
| BLANK / ZERO SETTING | Reagent |
| REAGENT VOLUME | 1 ml |
| SAMPLE VOLUME | 25 µl |
| INCUBATION TIME | 5 Minutes |
| STANDARD CONCENTRATION | 10 mg/dl |
| LOW NORMAL | 8.8 mg/dl |
| HIGH NORMAL | 10.2 mg/dl |
| LINEARITY | 16 mg/dl |

ASSAY PROCEDURE:

1. Take three clean, dry test tubes labeled B (blank), S (standard), T (test).
2. Set the instrument to zero with the blank, aspirate the standard to generate the factor.
3. Then aspirate the test sample one by one to read the result.

| | BLANK | STANDARD | SAMPLE |
|----------|-------|----------|--------|
| REAGENT | 1ml | 1ml | 1ml |
| STANDARD | | 25 µl | |
| SAMPLE | | | 25 µl |

Mix and read the optical density (A) after a 5-minute incubation at 37°C.

CALCULATIONS:

$$\text{Calcium Conc. (Mg/dl)} = \frac{\text{OD of Sample}}{\text{OD of Standard}} \times \text{Conc. of Standard}$$

LINEARITY:

Reagent is Linear up to 16 mg/dl.
Dilute the sample appropriately and re-assay if Calcium concentration exceeds 16 mg/dl. Multiply result with dilution factor.

REFERENCE NORMAL VALUE:

8.8-10.2 mg/dl

QUALITY CONTROL:

For accuracy it is necessary to run known controls with every assay.

LIMITATION & PRECAUTIONS:

1. Storage conditions as mentioned on the kit to be adhered.
2. Do not freeze or expose the reagents to higher temperature as it may affect the performance of the kit.
3. Before the assay bring all the reagents to room temperature.
4. Avoid contamination of the reagent during assay process.
5. Use clean glassware free from dust or debris.

BIBLIOGRAPHY:

Bishop, M L Dube-Von Laufen, J L., Burtis, Carl Aa and Ashwood, Titz 110, 61.