



CALCIUM Arsenazo



Arsenazum III colorimetric method for quantitative measurement of Calcium in serum, plasma and urine

ORDER INFORMATION

REF	Kit size
GA4323 00	10x50 ml
KL4323 00	6x60 ml
BK4323 00	2x60 ml

INDICATION

Conditions such as parathyroid disorders, neoplasms with or without bone metastasis, myelomas or other bone diseases can cause alterations in calcium levels.

METHOD PRINCIPLE

At neutral pH, Calcium present in the sample is complexed by Arsenazum III to form a coloured compound whose intensity, at 650 nm, is directly proportional to Calcium concentration in the sample.

COMPOSITION

REAGENT A:	
MES	90 mmol/l
Arsenazum III	0.25 mmol/l

STANDARD: 1x5 ml
 Calcium 10 mg/dl
 Verified against NIST reference material.

Preparation:

The reagents are liquids, ready to use.

Storage and stability

Store at room temperature (15-25 °C). Do not freeze the reagents! The reagents are stable up to the expiry date stated on the label.

After opening

REAGENT A and Standard are stable until the expiry date, if contamination and evaporation are avoided, protected from light. The above conditions are valid if the vials are opened just only for the time to take the reagent, closed immediately with their cap and stored at the indicated conservation temperature.

ANCILLARY EQUIPMENT

- Automatic pipettes
- Photometer
- Analysis cuvettes (optical path = 1 cm)
- Temperature controlled water bath
- NaCl solution 9 g/l

SAMPLES

Serum, heparin plasma or 24h urine. Do not use haemolysed samples. Do not utilize anticoagulants containing Calcium salts.
24 h urine, diluted 1:3.

Specimen collection / Preanalytical factors

It is recommended that specimen collection should be carried out in accordance with NCCLS Document H11-A3.

INTERNAL QUALITY CONTROL

It is recommended to use commercial Quality Control sera with known Calcium concentration. Check that the values obtained are within the reference range provided.

ANALYTICAL PROCEDURE

Allow the reagents to reach working temperature before using.

Pipette into disposable or well clean cuvettes:

	Blank	Standard	Sample
Reagent A	1000 µl	1000 µl	1000 µl
Distilled H ₂ O	25 µl	-	-
Standard	-	25 µl	-
Sample	-	-	25 µl

Mix and incubate for **5 minutes** at **37 °C**.
 Read the absorbance (A) of the standard and of the samples at **650 nm**.

CALCULATION OF RESULTS

Serum-plasma

$$\text{Calcium, mg/dl} = \frac{A \text{ sample}}{A \text{ standard}} \times 10$$

Urine

$$\text{Calcium, mg/dl} = \frac{A \text{ sample}}{A \text{ standard}} \times 10 \times 3$$

Conversion factor

$$\text{Calcium [mg/dl]} \times 0.2495 = \text{Calcium [mmol/l]}$$

REFERENCE VALUES

Serum-plasma: 8-10 mg/dl
 Urine 24h: 100-300 mg/24h

Each laboratory should establish reference ranges for its own patients population.

ANALYTICAL PERFORMANCES

Precision

Within-run and between-run coefficients of variation have been calculated on replicates of two samples at different Calcium concentrations. The obtained results are reported in the following tables:

Within-run				
Sample	n	Mean (mg/dl)	SD	%CV%
Serum # 1	10	9.6	0.24	2.51
Serum # 2	10	14.7	0.44	3.01

Between-run				
Sample	n	Mean (mg/dl)	SD	%CV
Serum # 1	10	9.4	0.25	2.63
Serum # 2	10	14.5	0.45	3.08

Linearity

The assay is linear up to 16 mg/dl.

Sensitivity

Test sensitivity, in terms of limit of detection, is 1 mg/dl.

Correlation

A correlation study comparing the present method and a commercial one gave the following results:

$$y = 0.9596x + 0.3313 \text{ mg/dl} \quad r = 0.9411$$

Interferences

Hemoglobin	> 250 mg/dl
Bilirubin	> 20 mg/dl
Triglycerides	> 1000 mg/dl

PRECAUTIONS IN USE

The reagents contain inactive components such as preservatives (Sodium azide or others), surfactants etc. The total concentration of these components is lower than the limits reported by 67/548/EEC and 88/379/EEC directives about classification, packaging and labelling of dangerous substances. However, the reagents should be handled with caution, avoiding swallowing and contact with skin, eyes and mucous membranes.

As calcium is an ubiquitous ion, essential precaution must be taken against accidental contamination.

Only use disposable materials.

The use of laboratory reagents according to good laboratory practice is recommended.

Waste Management

Please refer to local legal requirements.

BIBLIOGRAPHY

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