DIALAB Produktion und Vertrieb von chemisch-technischen Produkten und Laborinstrumenten Gesellschaft m.b.H.
IZ NOE-Sued, Hondastrasse, Objekt M55, 2351 Wr. Neudorf, Austria
Phone: +43 (0) 2236 660910-0, Fax: +43 (0) 2236 660910-30, e-mail: office@dialab.at

Calcium, Arsenazo

(en) English

Reagent with ATCS*

REF	Content		
D01372B	1 x	1 L	reagent
D01376	5 x	100 mL	reagent
D01375	5 x	50 mL	reagent
D01377	5 x	25 mL	reagent
D01378	5 x	10 mL	reagent
D59911	10 x	50 mL	reagent
D0412917	9 x	65 mL	reagent
DA0811	5 x	50 mL	reagent
DT1011	4 x	50 mL	reagent
DK0710	5 x	50 mL	reagent
DE1811	5 x	20 mL	reagent
DB20308	10 x	50 mL	reagent

* Advanced Turbidity Clearing System; minimizes turbidity caused by lipemia.

For professional in vitro diagnostic use only.

INTENDED USE

Diagnostic reagent for quantitative in vitro determination of calcium in human serum, plasma or urine on photometric systems.

DIAGNOSTIC SIGNIFICANCE[1,2]

Calcium plays an essential role in many cell functions: intracellularly in muscle contraction and glycogen metabolism, extracellularly, in bone mineralization, in blood coagulation and in transmission of nerve impulses. Calcium is present in plasma in three forms: free, bound to proteins or complexed with anions as phosphate, citrate and bicarbonate. Decreased total calcium levels can be associated with diseases of the bone apparatus (especially osteoporosis), kidney diseases (especially under dialysis), defective intestinal absorption and hypoparathyroidism. Increased total calcium can be measured in hyperparathyroidism, malignant diseases with metastases and sarcoidosis. Calcium measurements also help in monitoring of calcium supplementation mainly in the prevention of osteoporosis.

TEST PRINCIPLE

At neutral pH, calcium forms a blue coloured complex with arsenazo III. The intensity of the colour is proportional to the calcium concentration. Interference by magnesium is eliminated by addition of 8-hydroxyquinoline-5-sulfonic acid.

REAGENT COMPOSITION

COMPONENTS	CONCENTRATION	
Phosphate buffer, pH 7.5	50	mmol/L
8-Hydroxyquinoline-5-sulfonic acid	5	mmol/L
Arsenazo III	120	µmol/L

MATERIAL REQUIRED BUT NOT PROVIDED

- Standard or Calibrator, e.g.:

REF	Name	Content
D95094	Calcium Standard	1 x 3 mL
D98485	Diacal Auto	5 x 3 mL
D98485SV	Diacal Auto	1 x 3 mL

- Controls, e.g.:

REF	Name	Content	Description
D98481	Diacon N	12 x 5 mL	Control normal
D14481	Diacon N	5 x 5 mL	Control normal
D98481SV	Diacon N	1 x 5 mL	Control normal
D98482	Diacon P	12 x 5 mL	Control abnormal
D14482	Diacon P	5 x 5 mL	Control abnormal
D98482SV	Diacon P	1 x 5 mL	Control abnormal
D08581	Diacon Urine Level 1	12 x 5 mL	Urine Ctrl. normal
D08581SV	Diacon Urine Level 1	1 x 5 mL	Urine Ctrl. normal
D08582	Diacon Urine Level 2	12 x 5 mL	Urine Ctrl. abnormal
D08582SV	Diacon Urine Level 2	1 x 5 mL	Urine Ctrl. abnormal

- NaCl solution (9 g/L).
- Photometric device.
- General laboratory equipment.

REAGENT PREPARATION

The reagent is ready to use.

STORAGE AND STABILITY

Conditions:	Close immediately after use
	Avoid contamination
	Do not freeze the reagent
Storage:	at 2 – 8 °C
Stability:	up to the indicated expiration date

WARNINGS AND PRECAUTIONS

1. As calcium is an ubiquitous ion, essential precaution must be taken against accidental contamination. Only use disposable materials.
2. Traces of chelating agent, such as EDTA can prevent the formation of the coloured complex.
3. The reagents contain sodium azide (0.95 g/L) as preservative. Do not swallow! Avoid contact with skin and mucous membranes.
4. In very rare cases, samples of patients with gammopathy might give falsified results[7].
5. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents.
6. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
7. In the event of an incident related to the device, report it to the manufacturer and your competent authority as required.
8. For professional use only!

SPECIMEN COLLECTION AND STORAGE

Use serum, heparin plasma or urine.

Do not use EDTA plasma.

Sample preparation (Urine): add 10 ml of concentrated HCl to 24 h Urine and heat the specimen to dissolve calcium oxalate.

Stability[5]:

In serum/plasma	at 20 – 25 °C	7 days
	at 4 – 8 °C	3 weeks
	at - 20 °C	8 months
In urine:	at 20 – 25 °C	2 days
	at 4 – 8 °C	4 days
	at - 20 °C	3 weeks

Discard contaminated specimens. Freeze only once!

STANDARD

(Not included in the kit; has to be ordered separately)

Concentration:	10 mg/dL (2.5 mmol/L)
Storage:	2 – 8 °C
Stability:	up to the indicated expiration date

Protect from light! Close immediately after use!

TEST PROCEDURE

Method: Colorimetric, endpoint, increasing reaction, Arsenazo III

Wavelength: 650 nm, Hg 623 nm (630 – 670 nm)

Optical path: 1 cm

Temperature: 20 – 25 °C, 37 °C

Bring reagents and samples to room temperature.

Pipette into test tubes	Blank	Std./Cal.	Sample
Reagent	1000 µL	1000 µL	1000 µL
Sample	-	-	10 µL
Std./Cal.	-	10 µL	-
Dist. water	10 µL	-	-
Mix, incubate for 5 minutes at 20 – 25 °C / 37 °C and read absorbance against reagent blank.			

Automation

Special adaptations for automated analysers can be made on request.

INTERPRETATION OF RESULTS

Calculation

$$\text{Calcium [mg/dL]} = \frac{\Delta A \text{ Sample}}{\Delta A \text{ Std/Cal}} \times \text{conc. of Std/Cal [mg/dL]}$$

Unit Conversion

Calcium [mg/dL] x 0.2495 = Calcium [mmol/L]
Calcium (urine) [mg/24h] x 0.025 = Calcium (urine) [mmol/24h]

QUALITY CONTROL AND CALIBRATION

All control sera with calcium values determined by this method can be used.
We recommend the DIALAB serum controls **Diacon N** (control serum with values in the normal range) and **Diacon P** (control serum with values in the abnormal range) as well as the DIALAB urine controls **Diacon Urine Level 1** (control urine normal) and **Level 2** (control urine abnormal).
Each laboratory should establish corrective action in case of deviations in control recovery.

Calibration

The assay requires the use of a calcium standard or a calcium calibrator.
We recommend the DIALAB **Calcium Standard** and the DIALAB multi calibration serum **Diacal Auto.**

PERFORMANCE CHARACTERISTICS

Accuracy and precision

CV ≤ 1.73 % for within-run precision and CV ≤ 2.01 % for between-run precision.

Analytical sensitivity

The lower limit of detection is 0.04 mg/dL (0.01 mmol/L).

Linearity and measuring range

The test has been developed to determine calcium concentrations within a measuring range from 0.04 – 20 mg/dL (0.01 – 5 mmol/L). When values exceed this range samples should be diluted 1+1 with NaCl solution (9 g/L) and the results multiplied by 2.

Analytical specificity

no interference up to:

Ascorbic acid	30 mg/dL
Bilirubin	40 mg/dL
Hemoglobin	500 mg/dL
Triglycerides	2000 mg/dL
Magnesium	15 mg/dL

Strontium salts in medicine may lead to strongly increased calcium values.
For further information on interfering substances refer to Young DS[6].

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Clinical performance
A comparison of DIALAB Calcium, Arsenazo (y) with a commercially available assay (x) using 70 samples gave following results:
y = 1.02 x – 0.20 mg/dL; r = 0.999.

Tests were performed on the following instruments: Uvikon 922, Hitachi 704/911.

TRACEABILITY

This method has been standardized against the reference method Atomic Absorption Spectrometry (AAS).

EXPECTED VALUES

Serum/plasma[2]:	mg/dL	mmol/L
	8.6 – 10.3	2.15 – 2.57
Urine[1]:	mg/24h	mmol/24h
Women:	< 250	< 6.24
Men:	< 300	< 7.49

Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

WASTE MANAGEMENT

Please refer to local legal requirements.

LITERATURE

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7. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. ClinChemLabMed 2007; 45(9): 1240-1243.

USED SYMBOLS

Symbol	Description
Cont.	Content