CE IVD DIALAB

Liquid Reagents – ready to use

CALCIUM

CPC with ATCS*
2 Reagents

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

REF Cont.

D99097	**5 x 100 mL**	4 x 100 mL	Reagent 1
		1 x 100 mL	Reagent 2
D95098	**5 x 50 mL**	4 x 50 mL	Reagent 1
		1 x 50 mL	Reagent 2

Additionally offered:

D95094	1 x 3 mL	Calcium Standard	
D98485	5 x 3 mL	Calibrator	Diacal Auto
D98481	12 x 5 mL	Control normal	Diacon N
D98482	12 x 5 mL	Control abnormal	Diacon P
D08581	12 x 5 mL	Urine Ctrl. norm.	Diacon Urine Level 1
D08582	12 x 5 mL	Urine Ctrl. abnorm.	Diacon Urine Level 2

TEST PARAMETERS

Method:	Colorimetric, Endpoint, Increasing Reaction, CPC
Wavelength:	570 nm (550 - 590 nm), Hg 578 nm
Temperature:	20 – 25 °C, 37 °C
Sample:	Serum or heparinized plasma, acidified urine (do not use EDTA plasma)
Linearity:	up to 25 mg/dL (6.25 mmol/L) on Hitachi 911
Sensitivity:	The lower limit of detection is 0.2 mg/dL (0.05 mmol/L)

* **A**dvanced **T**urbidity **C**learing **S**ystem; minimzes turbidity caused by lipemia

REAGENT COMPOSITION

COMPONENTS		CONCENTRATION
Reagent 1	pH 10.7	
Ethanolamine	750	mmol/L
Detergents		
Reagent 2	pH 1.1	
o-Cresolphtalein complexone	0.13	mmol/L
8-Hydroxychinoline	35	mmol/L
Hydrochloric acid	100	mmol/L

REAGENT PREPARATION

Substrate Start:
Reagents are ready for use.

Sample Start:
Mix 4 parts of Reagent 1 with 1 part of Reagent 2.
(= Working Reagent)

REAGENT STABILITY AND STORAGE

Conditions: protect from light
close immediately after use, otherwise the pH decreases because of CO_2 absorption from the air.
do not freeze the reagents!

Substrate Start:
Storage: at 2 – 8°C
Stability: up to the expiration date

Sample Start (Working Reagent):
Stability in closed vials: at 2 – 8 °C 3 days
at 15 – 25°C 3 days

SAMPLE PREPARATION

Urine: add 10 mL of conc. HCl to 24 h Urine and heat the specimen to dissolve calcium oxalate

SAMPLE STABILITY AND STORAGE [5]

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

Discard contaminated specimens.

STANDARD

(has to be ordered separately)
Concentration: 10 mg/dL (2.5 mmol/L)
Storage: 2 – 25°C
Stability: up to the expiration date
CLOSE IMMEDIATELY AFTER USE!

INTERFERING SUBSTANCES

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.

MANUAL TEST PROCEDURE

Bring reagents and samples to room temperature.

Note: For measurement of coloured or lipemic samples use substrate start.

Substrate Start:

Pipette into test tubes	Blank	Std./Cal.	Sample
Sample	-	-	20 µL
Std./Cal.	-	20 µL	-
Dist. water	20 µL	-	-
Reagent 1	1000 µL	1000 µL	1000 µL
Mix and read absorbance A1 against reagent blank after 5 - 30 min. at 20-25°C/37°C. Then add:			
Reagent 2	250 µL	250 µL	250 µL
Mix and read absorbance A2 against reagent blank after 5 - 30 min. at 20-25°C/37°C ΔA = (A2-A1) sample or Std./Cal.			

Sample Start:

Pipette into test tubes	Blank	Std./Cal	Sample
Sample	-	-	20 µl
Std./Cal.	-	20 µl	-
Dist. water	20 µl	-	-
Working Reagent	1000 µl	1000 µl	1000 µl
Mix and read absorbance against reagent blank after 5 - 30 min at 20-25°C/37°C.			

CALCULATION (light path 1 cm)

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

UNIT CONVERSION

mg/dL x 0.2495 = mmol/L

REFERENCE RANGE [2] *

serum/plasma:	mg/dL	mmol/L
	8.6 - 10.3	2.15 – 2.57

urine:	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.

TEST PRINCIPLE

Cresolphtalein complexone (CPC) reacts with Calcium ions in alkaline solution forming a violet colour.
The intensity of the violet color is proportional to the calcium concentration in the sample.
Interference by magnesium is eliminated by addition of 8-hydroxy-quinoline.

PERFORMANCE CHARACTERISTICS

LINEARITY

The assay is linear between 0.2 - 25 mg/dL (0.05 – 6.25 mmol/L) on Hitachi 911. Above this concentration, samples should be diluted 1 + 1 with NaCl solution (9 g/L) and reassayed multiplying the result by 2.

PRECISION (at 37°C)

Intra-assay n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	6.18	0.05	0.84
Sample 2	9.94	0.10	1.02
Sample 3	13.5	0.11	0.81

Inter-assay n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	6.31	0.09	1.38
Sample 2	10.1	0.10	1.04
Sample 3	13.4	0.08	0.63

METHOD COMPARISON

A comparison between Dialab Calcium (y) and a commercially available test (x) using 82 samples gave following results:
y = 0.98 x + 0.11; r= 0.999.

QUALITY CONTROL

All controls with Calcium values determined by this method can be used.

We recommend:

REF Cont.

D98481	12 x 5 mL	**DIACON N**	Assayed Ctrl Serum Norm.
D98482	12 x 5 mL	**DIACON P**	Assayed Ctrl. Serum Abnorm.
D08581	12 x 5 mL	**Diacon Urine Level 1**	Urine Ctrl. normal
D08582	12 x 5 mL	**Diacon Urine Level 2**	Urine Ctrl. abnorm.

CALIBRATION

The assay requires the use of a Calcium Standard or a Calcium Calibrator.
We recommend:

REF Cont.

D95094	1 x 3 ml	**CALCIUM STANDARD**	
D98485	5 x 3 ml	**DIACAL AUTO**	Assayed Multi Calibration Serum

AUTOMATION

Special adaptations for automated analyzers can be made on request.

WARNINGS AND PRECAUTIONS

1. Reagent 1 is irritating: Xi
   R36: Irritating to eyes.
   S2: keep out of the reach of children.
   S25: Avoid contact with eyes.
   S26: In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
2. Reagent 2:
   S24/25: Avoid contact with skin and eyes.
3. As calcium is an ubiquitous ion, essential precaution must be taken against accidental contamination. Only use disposable materials.
4. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents.

WASTE MANAGEMENT

Please refer to local legal requirements.

REFERENCES

1. Thomas L., Clinical Laboratory Diagnostics, 1st ed. Frankfurt: TH-Books Verlagsgesellschaft; 1998, p. 192-202.
2. Endres DB, Rude R.K., Mineral and bone metabolism, In: Burtis CA, Ashwood ER, editors Tietz Textbook of Clinical Chemistry 3rd ed. Philadelphia: W.B. Saunders Company; 1999, p. 1395-1457.
3. Baginski E.S., Marie S.S., Clark W.L., Zak B. Direct microdetermination of serum calcium. Clin Chim Acta 1973; 46: 46-54.
4. Sarkar BCR, Chauhan UPS. A new method of determining micro quantities of calcium in biological materials. Anal. Biochem. 1967; 20: 155-166.
5. Guder WG, Zawta B et al. The quality of Diagnostic Samples. 1st ed. Darmstadt: GIT Verlag; 2001; p.20-1, 50-1.

2°C 8°C IVD

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