

CALCIUM REAGENT KIT

(ARSENAZO III METHOD)

Quantitative determination of calcium in Serum Only for In Vitro diagnostic use

Ref No.

Ca 50 Ca 100

Summary

Calcium, in the body, is found mainly in the bones. In serum calcium exists equally in a free ionized form and in a bound form(with albumin). Hence a decrease in albumin cause lower calcium levels & vice versa. The levels of calcium in serum depend on the parathyroid hormone. Increased calcium level are found in bone tumours, hyperparathyroidism. Decreased levels are found in hypoparathyroidism, renal failure, rickets, Vitamin D deficiency & pancreatitis.

Principle

Calcium combines specifically with Arsenazo 111 at a neutral pH to form a blue purple colored complex. The intensity of the developed color is proportional to the calcium ion concentration of the sample.

Calcium + Arsenazo III ----> Blue purple colored complex

Kit contents		
Kit size	50ml	100ml
Ref no.	Ca50	Ca100
Calcium	2	2
Reagent		
Calcium	1	1
Standard		
IFU	1	1

Material required but not provided

Test tubes, yellow tips, blue tips, Pipetting devices, Timing device and heating block to maintain temperature.

Storage& Stability of the Reagents

1. The reagents are stable up to the expiry date labeled on the bottles when stored at 18C-30°C, protect from light &contamination is avoided.

- 2. Do not freeze the reagents.
- 3. Ensure the specimens are brought to Room Temperature.
- 4. Ensure the reagents shelf life is valid.
- 5. Do not use haemolysed & lipemic serum.

Reagent Preparation

The reagent and standard are ready to use

Reagent Composition

nt1	Pipes buffer pH -6.8	15.2g/l
	Arsenazo dye	0.052g/l
	Potassium ferrocyanide	0.18g/l
	DMSO	20ml/1
	Sodium azide	0.28g/l
ırd	Calcium	10mg/dl
	nt1 ard	t1 Pipes buffer pH -6.8 Arsenazo dye Potassium ferrocyanide DMSO Sodium azide rd Calcium

Specimen

Serum,

Specimen collection

- 1. Fresh, clear, non-hemolysed serum from fasting patients is recommended.
- 2. EDTA, sodium fluoride, sodium citrate and oxalate should be avoided because they interfere with the results.

Storage & Stability of the Specimen

7days	at	15°C-25°C
22days	at	$2^{0}C-8^{0}C$
Discard cont	aminat	ed specimens.

Warning & Precautions

- 1. Keep out of reach children. In case of contact with eyes, rinse immediately with plenty of water &seek medical advice.
- 2. Take off immediately all contaminated clothing.
- 3. Wear suitable gloves and eye /face protection.
- 4. Do not pipette by mouth. Always use safety pipettes to pull the reagents.
- 5. Reagents may contain some non-reactive and preservative components. It is suggested to handle carefully, avoid direct contact with skin and do not swallow.
- 6. Perform the test according to the current "Good Laboratory Practice" (GLP) guidelines.

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7. The reagents contain sodium azide (0.95g/L) as preservative. Do not swallow. Avoid contact with skin and mucous membrane.

Assay Procedure

Wave length		630 nm		
Temperatur	re :	$37^{0} c$		
Light path		: 10 mm		
Measureme	ent	: against reagent blank		
	Blank	Standard	Test	
Reagent	1000µ1	1000µ1	1000 <mark>µ1</mark>	
-				
Sample/				
Standard	-	25µ1	25μ l	

Mix & incubate for 10minutes at Room temperature, then read the absorbance of sample and standard against reagent blank at 630 nm.

Calculations:

Abs. of sample

Abs. of Std.

X 10= Conc. of calcium in mg/dl

Linearity

The linearity is 16.0mg/dl.

Note

As calcium is a very widely distributed ion, care should be taken to avoid any contamination. All glassware being used for the test should first be rinsed with 1% or 0.1 N HCL & then with the good quality deionised water before use.

Reference Range

Serum/Plasma

"Each laboratory should check if references ranges are transferable to its own patient population & determine own preference ranges if necessary".

8.7-11mg/dl

Quick References

Parameter	Calcium
Mode	Endpoint
Wavelength	630nm
Unit	mg/dl
Temperature	37°C
Standard conc.	10mg/dl
Reaction slope	Increasing
Reagent volume	1000µ1
Sample volume	25µ1
Incubation time	10mins.at RT
Blanking	Reagent blank
linearity	16mg/dl

References:

- 1. Faulker, W.R. and Meites, S.: Selected Methods for the Small clinical chemistry Laboratory.P. 125 Washington, D.C. 1982.
- 2. Gitelman, h.: Anal. Biochem. 20:5521 (1967).

Note on symbols and marks

I		LOT	
Instructions for use	Use by	Batch number	Manufacturer
IVD Invitro Diagnostic Medical Device	Date of manufacturer	Temperature limit	REF Reference number