BIOLABO
www.biolabo.fr

MANUFACTURER:
BIOLABO SAS,
Les Hautes Rives
02160, Maizy, France

MAGNESIUM Calmagite

Reagent for quantitative determination of magnesium in human serum, plasma, red blood cells (RBC) and urines.

REF 87212 **R1** 2 x 250 mL **R2** 1 x 10 mL **R3** 1 x 15 mL **R4** 1 x 15 mL **R5** 1 x 10 mL

TECHNICAL SUPPORT AND ORDERS
Tél : (33) 03 23 25 15 50
Fax: (33) 03 23 256 256

CE

IVD IN VITRO DIAGNOSTIC USE

CLINICAL SIGNIFICANCE (1)

Adult human body (70 Kg) contains 21 to 28 g of magnesium. Of this, about 60% is in bone, 20% in skeletal muscle, 19% in other cells, and about 1% in the extracellular fluids. About 30% of magnesium in plasma is associated with proteins (primarily albumin). Consequently, a change in the concentration in albumin can affect the concentration in magnesium.
Hypomagnesaemia may be a secondary effect in hypocalcemic or calcium-deficient tetany. Conditions that have been associated with hypomagnesemia include chronic alcoholism, childhood malnutrition, lactation, malabsorption, acute pancreatitis, hypothyroïdism, chronic glomerulonephritis, aldosteronism, digitalis intoxication and prolonged intravenous feeding.
Hypermagnesaemia have been observed in dehydratation, severe diabetic acidosis, and immediately following myocardial infarction.

PRINCIPLE (1) (4) (5)

Gindler, Heth and Khayam-Bashi method. Calmagite, a metallochromic indicator (1 -[1-hydroxy-4-methyl-2-phenylazo]-2-naphtol-4-sulfonic acid), forms a coloured complex with the magnesium in basic medium. The absorbance, measured at 510-550 nm, is proportional to the concentration of magnesium in the specimen. EGTA reduces Calcium interference, Potassium cyanide (KCN) reduces interference of heavy metals, Polyvinylpyrolidone (PVP) and surfactants reduce the interference of proteins and lipemia.

REAGENTS COMPOSITION

Vial R1 **CALMAGITE REAGENT**

Calmagite ≥ 223 µmol/L KCN 6.14 mmol/L PVP 40 g/L
KOH 38 mmol/L EGTA 250 µmol/L Surfactants

Vial R2 **STANDARD**

Magnesium 2 mg/dL (0.822 mmol/L)

Vial R3 **PRECIPITANT**

Sodium tungstate 300 mmol/L

Vial R4 **SULFURIC ACID**

H_2SO_4 350 mmol/L

Xi, R36/38: Irritating to eyes and skin.
S26-S28: After contact with eyes, rinse immediately with plenty of water and seek medical advice. After contact with skin, rinse immediately with plenty of water.
S36/37/39: Wear suitable protective clothing, gloves and eyes/face protection

Vial R5 **STANDARD**

Magnesium 6 mg/dL (2.47 mmol/L)

SAFETY CAUTIONS

BIOLABO reagents are designated for professional, in vitro diagnostic use.

- Verify the integrity of the contents before use.
- Use adequate protections (overall, gloves, glasses).
- Do not pipette by mouth.
- In case of contact with skin or eyes wash affected areas with plenty of water and seek medical advice.
- Material Safety Data Sheet is available upon request.
- Waste disposal: Respect legislation in force in the country.

All specimens should be handled as potentially infectious, in accordance with good laboratory practices using appropriate precautions. Respect legislation in force in the country.

REAGENTS PREPARATION

Reagents are ready for use.

STABILITY AND STORAGE

Store away from air and light, well cap in the original vial at 18-25°C

- Standards (vial R2, R5): Transfer the requested quantity, recap and store at 18-25°C.
- Contents of vial R1: transfer the requested quantity, promptly recap and store at 18-25°C.
- Without contamination, reagents are stable until expiry date stated on the label of the kit when stored and used as described in the insert.
- Discard any reagent if cloudy and reagent (vial R1) if its absorbance measured at 530 nm < 0.600.

SPECIMEN COLLECTION AND HANDLING

Collect in a metal-free container and without preservatives
Unhemolysed serum or heparinised plasma: Collect on fasting. Avoid oxalate, citrate or EDTA. Separate red cells immediately.
Special procedure is required for cloudy or icteric serum (see next page).

Magnesium is stable for several days in serum at 2-8°C.

24h urines (acidified pH 1.0): dilute (1+4) with demineralised water before assay.

RBC: Collect venous blood on heparin and centrifuge. After elimination of plasma, wash RBC 3 times with saline solution, then centrifuge 10 minutes at 4000 RPM and eliminate the supernatant.

INTERFERENCES (3)

- Give a special care to the specimen, calibrators and controls handling, to avoid contamination by the environmental magnesium. The use of disposable tubes or cuvettes and acid washed labware (well rinsed with demineralised water) is suggested.
- Calcium (≤ 7.5 mmol/L) does not interfere with this method.
- Plasma, serum: Icterus, lipemia and paraproteins may interfere with the determination. Hemolysis involves an overestimation because of the important intracellular contents in magnesium.

For a more comprehensive review of factors affecting this assay refer to the publication of Young D.S.

CALIBRATION (6)

- Standards (vial R2 or R5) enclosed in the kit or BIOLABO Multicalibrator REF 95015 traceable to SRM 909b.
- Or any calibrator traceable to a reference method or material.

The calibration frequency depends on proper instrument functions and on preservation of the reagent.
It is recommended to calibrate in the following cases:
1. When changing batch of reagent.
2. After maintenance operations on the instrument .
3. When control values obtained are out of range, even after using a new vial of fresh serum

Manufacturer | Use by | IVD In vitro diagnostic | Temperature limitation | REF Catalogue number | See insert | LOT Batch number | Store away from light | sufficient for | → dilute with

MATERIAL REQUIRED BUT NOT PROVIDED

1. Basic medical analysis laboratory equipment.
2. Normal and pathological control sera.

QUALITY CONTROL

- BIOLABO EXATROL-N Level I REF 95010.
- BIOLABO EXATROL-P Level II REF 95011.
- Assayed control sera referring to the same method.
- External quality control program.

It is recommended to control in the following cases:

- At least once a run.
- At least once within 24 hours.
- When changing vial of reagent.
- After maintenance operations on the instrument.

If control is out of range, apply following actions:

1. Repeat the test with the same control.
2. If control is still out of range, prepare a fresh control serum and repeat the test.
3. If control is still out of range, use a new vial of calibrator or a fresh calibrator and repeat the test.
4. If control is still out of range, calibrate with a new vial of reagent.
5. If control is still out of range, please contact BIOLABO technical support or your local Agent.

EXPECTED VALUES (2)

Serum or Plasma	mg/dL	[mmol/L]
Newborn	1.5-2.2	[0.62-0.91]
Child	1.7-2.2	[0.70-0.91]
Adult	1.6-2.6	[0.66-1.07]
RBC	4.01-6.44	[1.65-2.65]
Urines	73-122 mg/24h	[3.00-5.00 mmol/24 h]

Each laboratory should establish its own normal ranges for the population that it serves.

LINEARITY

Procedure n°2: linear up to 8.0 mg/dL (3.29 mmol/L).
Procedure n°1 and n°3: linear up to 4.0 mg/dL (1.65 mmol/L)

Above, dilute the specimen with saline solution and reassay taking into account dilution factor. Linearity limit depends on specimen/reagent ratio.

PERFORMANCES

Procedure n°1:

Within run N = 20	*Medium level*	*High level*
Mean mg/dL	2.42	3.71
S.D. mg/dL	0.03	0.05
C.V. %	1.2	1.3

Between run N > 20	*Medium level*	*High level*
Mean mg/dL	2.07	3.47
S.D. mg/dL	0.056	0.104
C.V. %	2.7	3.0

Detection limit: approximately 0.15 mg/dL

Sensitivity for 2 mg/dL: approximately 0.154 Abs at 530 nm.

Comparison study with commercially available reagent:

y = 0.9614 x + 0.02671 r =0.9953

CALCULATION

Calculate the result as follows:

Serum, plasma and RBC:

$$\text{Result} = \frac{\text{Abs (Assay)}}{\text{Abs (Standard)}} \times \text{Standard concentration}$$

Urines diluted (1+4): multiply the result by 5 (dilution factor).

REFERENCES

(1) TIETZ N.W. Text book of clinical chemistry, 3rd Ed. C.A. Burtis, E.R. Ashwood, W.B. Saunders (1999) p.1034-1036 et 1408-1410.
(2) Clinical Guide to Laboratory Test, 3rd Ed., N.W. TIETZ (1995) p. 418-420.
(3) YOUNG D.S., Effect of Drugs on Clinical laboratory Tests, 4th Ed. (1995) p. 3-410 to 3-414
(4) GINDLER E.M., HETH D.A., Clin. Chem. (1971), 17, p.662
(5) H.KHAYAM-BASHI, TSAN Z. LIU, VERN W. Clin. Chem. (1977), 23/2, p.289-291
(6) SRM: Standard Reference Material ®

MANUAL PROCEDURE

In any case:

- Let stand reagents and specimens at room temperature
- Maintain a constant temperature as the reaction is temperature sensitive.
- Reaction is stable for 60 minutes.

Procedure n°1: Clear specimen (note 1)

Pipette into well identified test tubes:	Blank	Standard	Assay
Reagent R1	1 mL	1 mL	1 mL
Demineralised water	10 µL		
Standard R2 (2 mg/dL)		10 µL	
Specimen (Note 1)			10 µL
Mix. Let stand for 5 minutes at constant temperature. Read standard and assays absorbances at 530 nm (510-550) against reagent blank.			

Notes:

1. Serum, plasma or urines diluted (1+4) with demineralised water.
2. Specific procedures are available upon request for automated instruments. Please contact BIOLABO technical support.

Procedure n°2: RBC
(see § **SPECIMEN COLLECTION AND HANDLING**)

Pipette into well identified test tubes:	Blank	Standard	Assay
Demineralised water	3.2 mL	3 mL	3 mL
Standard R5 (6 mg/dL)		200 µL	
Globular clot (RBC)			200 µL
Mix vigorously until complete hemolysis. It is recommended to use a positive moved pipette to dispense globular clot. Well rinse pipette tip by movement to and fro.			
Vial R3 (Precipitant)	400 µL	400 µL	400 µL
Mix			
Vial R4 (H_2SO_4)	400 µL	400 µL	400 µL
Mix well. Wait for 5 minutes, centrifuge at 4000 RPM for 10 minutes. Pipette into identified tubes			
Reagent R1	1 mL	1 mL	1 mL
Supernatant	100 µL	100 µL	100 µL
Mix. Let stand for 5 minutes at constant temperature. Read standard and assays absorbances at 530 nm (510-550) against reagent blank.			

Procedure n°3: Cloudy or icteric sera

Pipette into well identified test tubes:	Blank	Standard	Assay
Serum			200 µL
Standard R2 (2 mg/dL)		200 µL	
Demineralised water	200 µL		
Vial R3 (Precipitant)	400 µL	400 µL	400 µL
Mix			
Vial R4 (H_2SO_4)	400 µL	400 µL	400 µL
Mix well. Let stand for 5 minutes and centrifuge at 4000 RPM for 10 minutes. Pipette into well identified test tubes:			
Reagent R1	1 mL	1 mL	1 mL
Supernatant	50 µL	50 µL	50 µL
Mix. Let stand for 5 minutes at constant temperature. Read standard and assays absorbances at 530 nm (510-550) against reagent blank.			