PRODUCT INFORMATION & MANUAL

Magnesium Assay Kit (Colorimetric)

NBP3-25897

For research use only.
Not for diagnostic or therapeutic procedures.

www.novusbio.com - P: 303.730.1950 - P: 888.506.6887 - F: 303.730.1966 - technical@novusbio.com

Novus kits are guaranteed for 6 months from date of receipt

Magnesium (Mg) Colorimetric Assay Kit

Catalog No: NBP3-25897

Method: Colorimetric method

Specification: 100Assays (Can detect 96 samples without duplication)

Measuring instrument: Spectrophotometer

Sensitivity: 0.12 mmol/L

Detection range: 0.12-2.50 mmol/L

Average intra-assay CV (%): 4.8

Average inter-assay CV (%): 9.9

Average recovery rate (%): 95

- This kit is for research use only.
- Instructions should be followed strictly, changes of operation may result in unreliable results.
- Please kindly provide us the lot number (on the outside of the box) of the kit for more efficient service.

General information

▲Intended use

The kit can be used to detect concentration of magnesium (Mg) in plasma or serum samples.

▲Background

Magnesium is an important biological element, which is mainly found in bone, muscle cells, soft tissues, serum and red blood cells. It is involved in the synthesis of nucleic acid and protein, and is a cofactor of various enzymes and transporters. It plays an important role in regulating cardiac excitability, neuromuscular conduction, vasomotor contraction, blood pressure and energy metabolism.

▲Detection principle

The magnesium in the serum reacts with the complexometric indicator (Calmagite) to form the Calmagite-Mg compound. The absorbance of this compound at 540 nm is proportional to the concentration of magnesium in the sample. The concentration of magnesium can be calculated by measuring the OD value at 540 nm.

▲Kit components & storage

Item	Component	Specification	Storage
Reagent 1	Alkali Reagent	60 mL × 1 vial	2-8℃ , 12 months
Reagent 2	Chromogenic Agent	60 mL × 1 vial	2-8℃ , 12 months, shading light
Reagent 3	5 mmol/L Magnesium Standard	1 mL × 2 vials	2-8℃ , 12 months
Note: The reagents must be stored strictly according to the preservation conditions in the above table. The reagents in different kits cannot be mixed with each other.			

▲Materials prepared by users

Instruments

Spectrophotometer (540 nm), Micropipettor, Centrifuge, Incubator, Vortex mixer

Reagents:

Double distilled water, Normal saline (0.9% NaCl), PBS (0.01 M, pH 7.4)

⚠ Safety data

Some of the reagents in the kit contain dangerous substances. It should be avoided to touch the skin and clothing. Wash immediately with plenty of water if touching it carelessly. All the samples and waste material should be treated according to the relevant rules of laboratory's biosafety.

⚠ Precautions

Before the experiment, please read the instructions carefully, and wear gloves and work clothes.

▲ The key points of the assay

1. Prepare and store the working solution with shading light.
2. The assay temperature of this method is not required strictly. But it should be kept constant, because the color is sensitive to the temperature.
3. The color of reaction solution can be stable for 1 hour.
4. Plasma samples should be anticoagulant with heparin.

Pre-assay preparation

▲ Reagent preparation

The preparation of working solution

Mix the reagent 1 and reagent 2 at the ratio of 1:1 fully and stand for 10 min to prepare the working solution. The prepared solution can be stored at 2-8℃ with shading light for 3 days.

The preparation of 1 mmol/L standard solution

Dilute the reagent 3 with double distilled water for 5 times. The prepared solution can be stored at 2-8℃ with shading light for 3 days.

▲ Sample preparation

Sample requirements

Citrate and EDTA should not be used as anticoagulants.

Serum:

Collect fresh blood and stand at 25℃ for 30 min to clot the blood. Then centrifuge at 2000 g for 15 min at 4℃ . Take the serum (which is the upper light yellow clarified liquid layer) to preserve it on ice for detection. If not detected on the same day, the serum can be stored at -80℃ for a month.

Plasma:

Take fresh blood into the tube which has anticoagulant (heparin is used as anticoagulant, do not use citrate and EDTA as anticoagulants), centrifuge at 700-1000 g for 10 min at 4℃ . Take the plasma (which is the upper light yellow clarified liquid layer, don't take white blood cells and platelets in the middle layer) to preserve it on ice for detection. If not detected on the same day, the plasma can be stored at -80℃ for a month.

▲ Dilution of sample

It is recommended to take 2~3 samples with expected large difference to do pre-experiment before formal experiment and dilute the sample according to the result of the pre-experiment and the detection range (0.12-2.50 mmol/L).

The recommended dilution factor for different samples is as follows (for reference only):

Sample type	Dilution factor
Human serum	1
Rat serum	1
Mouse serum	1
Porcine serum	1
Chicken serum	1
Dog serum	1

Note: The diluent is normal saline (0.9% NaCl) or PBS (0.01 M, pH 7.4).

Assay protocol

▲Detailed operating steps

1. Blank tube: Add 1000 μL of working solution to 2 mL EP tube.
   Standard tube: Add 1000 μL of working solution to 2 mL EP tube.
   Sample tube: Add 1000 μL of working solution to 2 mL EP tube.
2. Incubate the tubes at 37℃ for 5 min.
3. Blank tube: Add 10 μL of double distilled water to blank tube.
   Standard tube: Add 10 μL of 1 mmol/L magnesium standard to standard tube.
   Sample tube: Add 10 μL of sample to sample tube.
4. Mix fully with vortex mixer and incubate the tubes at 37℃ for 2 min.
5. Set the spectrophotometer to zero with double distilled water and measure the OD value of each tube at 540 nm with 0.5 cm optical path quartz cuvette.

▲Summary operation table

	Blank tube	Standard tube	Sample tube
Working solution (μL)	1000	1000	1000
Incubation at 37℃ for 5 min.			
Double distilled water (μL)	10		
1 mmol/L Mg standard solution (μL)		10	
Sample (μL)			10
Mix fully and incubate the tubes at 37℃ for 2 min. Set the spectrophotometer to zero with double distilled water and measure the OD value at 540 nm.			

▲Calculation

$$\text{Mg content (mmol/L)} = \frac{\Delta A_1}{\Delta A_2} \times c \times f$$

Note:

ΔA_1: $OD_{Sample} - OD_{Blank}$

ΔA_2: $OD_{Standard} - OD_{Blank}$

c: The concentration of standard, 1 mmol/L.

f: Dilution factor of sample before tested.

Appendix I Data

▲Example analysis

Take 10 μL of mouse serum and carry the assay according to the operation table. The results are as follows:

The average OD value of the sample is 0.413, the average OD value of the blank is 0.309, the average OD value of the standard is 0.392, and the calculation result is:

$$\text{Mg content (mmol/L)} = \frac{0.413-0.309}{0.392-0.309} \times 1.0 = 1.25(\text{mmol/L})$$

Appendix II References

1. Mooren, C. F. Magnesium and disturbances in carbohydrate metabolism[J]. Diabetes Obesity & Metabolism, 2015, 17(9): 813-823.
2. Won S J, Jin P T. Magnesium Metabolism[J]. Electrolytes & Blood Pressure E & Bp, 2008, 6(2): 86-95.
3. Champagne C M. Magnesium in Hypertension, Cardiovascular Disease, Metabolic Syndrome, and Other Conditions: A Review[J]. Nutrition in Clinical Practice, 2008, 23(2): 142-151.
4. Noronha L J, Matuschak G M. Magnesium in critical illness: metabolism, assessment, and treatment[J]. Intensive Care Med, 2002, 28(6): 667-679.