Magnesium

Colorimetric Microplate Assay Kit

User Manual

Catalog # CAK1107

(Version 1.3C)

Detection and Quantification ofMagnesium(Mg^{2+}) ContentinSerum, Urine, Saliva and Other biological fluidsSamples.

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

I. INTRODUCTION

Magnesium (Mg) is one of the most abundant and essential minerals in mammals. Magnesium is involved in more than 300 biochemical reactions in the body and plays important roles in muscle and nerve functions, heart rhythm, immune system and bone formation. Magnesium deficiency may lead to nausea, fatigue, muscle contractions, hypocalcemia and hypokalemia.

The magnesiums can react with calmagite.The productscan be measured at a colorimetric readout at 520 nm.

II.KIT COMPONENTS

Component	Volume	Storage
96-Well Microplate	1 plate	
Dye Reagent I	Powderx 1	4 °C, keep in dark
Dye Reagent II	11 mlx 1	4 °C, keep in dark
Dye Reagent III	Powderx 1	4 °C
Reaction Buffer	5 mlx 1	4 °C
Standard (10mmol/L)	1 ml x 1	4 °C
Technical Manual	1 Manual	

III. MATERIALS REQUIRED BUT NOT PROVIDED

1. Microplate reader to read absorbance at 520 nm
2. Distilled water
3. Pipettor, multi-channel pipettor
4. Pipette tips
5. Mortar
6. Centrifuge
7. Timer

IV. REAGENT PREPARATION

Standard:Briefly centrifuge prior to opening.Perform 2-fold serial dilutions of the top standardsolution using distilled waterto make the standard curve.The concentration of standard curve could be10/5/2.5/1.25/0.625/0.312/0.156 mmol/L.

Dye Reagent I:Briefly centrifuge prior to opening. Add 1.5 ml distilled waterto dissolve before use. Keep in dark and store at -20 °C for 1-2weeks.

Dye Reagent III:Briefly centrifuge prior to opening. Add 1.5 ml distilled waterto dissolve before use.Store at 4°C for 5-6months.

Note: Divide into small aliquots to avoid repeated freeze-thaw cycles.

V. SAMPLE PREPARATION

1.For serumand other biological fluidssample

Detect directly.

VI. ASSAY PROCEDURE

Add following reagents intothe microplate:

Reagent*	Blank	Standard	Sample**
Reaction Buffer	50 µl	50 µl	50 µl
Dye Reagent I	15µl	15µl	15µl
Dye Reagent II	110 µl	110 µl	110 µl
Dye Reagent III	15µl	15µl	15µl
Mix well.			
Distilled water	10µl	--	--
Standard	--	10µl	--
Sample	--	--	10µl
Mix, measured at 520 nm and recordthe absorbance.			

Note:

*Reagents must be added sequentially and should not be premixed prior to addition.

**The concentrations can vary over a wide range depending on the different samples. For unknown samples, we recommend doing a pilot experiment & testing several doses to ensure the readings are within the standard curve range.

**For colored samples, we recommend setting a parallel sample background control well with same volume of sample only. Other reagents were replaced by distilled water to the same total volume. Subtract the OD value of the sample background control from the OD value of the sample to correct for interference from the sample's own color.

VII. CALCULATION

1. Calculatethe sample concentration in ASSAY PROCEDURE according to the slope of the standard curve

$$C=\frac{(OD_{Sample}-OD_{Blank})-\text{Intercept}}{\text{Slope}}\times n(mmol/L)$$

Calculate the initial concentration according to sample preparation procedure.

2. According to one point of the standard OD and concentration

According to the volume of sample

$$C=\frac{(C_{Standard}\times V_{Standard})\times(OD_{Sample}-OD_{Blank})}{(OD_{Standard}-OD_{Blank})\times V_{Sample}}(\mu mol/ml)$$

Slope: the absorbance slope of standard curve

n: the dilution factor

$C_{Standard}$: the standard concentration, mmol/L = μmol/ml

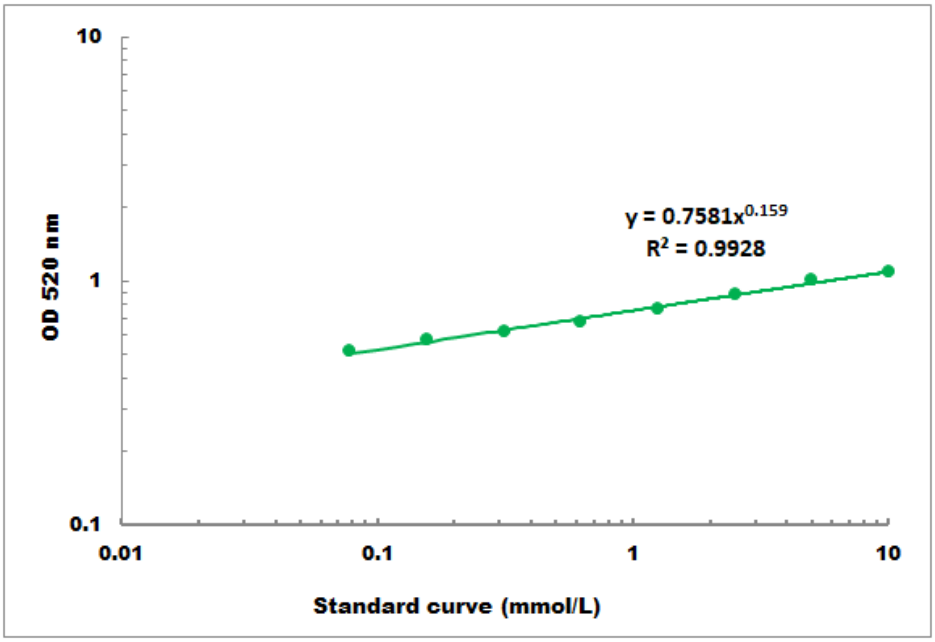
$V_{Standard}$: the volume of standard in assay procedure, μl

V_{Sample}: the volume of sample in assay procedure, μl

V_{Assay}: the volume of Assay Buffer, μl

VIII. TYPICAL DATA

The standard curve is for demonstration only. A standard curve must be run with each assay.



Detection Range: 0.1 mmol/L - 10 mmol/L