

Malachite Green Phosphate Colorimetric Assay Kit (TBS2042, 1000 Assays, Store at -20 °C)

Description

Phosphate is one of the most abundant minerals in the human body and other sources. Disruption of phosphate homeostasis was involved in many disease pathology processes through effected energy metabolism, cell signaling, regulation of protein synthesis. Malachite green complex is a useful dye for the measurement of free inorganic phosphate (Pi) from protein, lipid phosphorylation and other sources.

Tribioscience's Malachite Green Phosphate Kit provides a simple, easy assay that relies on quantification of free phosphate with malachite green complex. In this assay, the free phosphate combines malachite green reagent A and B to form color complex. The color intensity is quantified at 620 nm.

Applications

Measure phosphate in a variety of samples.

Kit Contents for 1000 tests:

Name	Part Size
Phosphate Standard (200 μM)	1 mL
Malachite Green Reagent A	12 ml
Malachite Green Reagent B	6 mL

Store the Reagent at -20°C, protect from light, for 12 months.

Sample Preparation

1. Samples should be diluted with ddH₂O.
2. Aliquot and store lysates at -80°C unless being used immediately.

Assay Procedures

Note: Equilibrate all the kit components until room temperature before starting the experiment.

1. Prepare the Phosphate standards: Label Tube number #1 to #8. Add 100 μL of dd H₂O from Tube #1 through to #7, add 100μL of dd H₂O to #8.
2. Make a 2-fold serial dilution by adding 100 μL of phosphate standard to the next concentration tube to make standard concentration from 100 to 1.56 μM shown in Table 1. Tube#8 as standard 0.

Table 1: Phosphate Standard Preparation

Tube	Pi standard Vol.(μL)	dd H ₂ O (μL)	Standard (μM)
1	100 μL of 200 μM	100	100
2	100 μL of 100 μM	100	50
3	100 μL of 50 μM	100	25
4	100 μL of 25 μM	100	12.5
5	100 μL of 12.5 μM	100	6.25
6	100 μL of 6.25 μM	100	3.125
7	100 μL of 3.125 μM	100	1.5625
8	0	100	0

3. Add 25 μL standards and samples in the appropriate wells.
4. Add 10 μL of Malachite Reagent A in each well. Then add 5 μL Malachite Reagent B in each well. And 10 μL dd-water to each well. Gently mixed thoroughly.

5. Incubate 5-10 min at room temperature.
6. Read plate at 620 nm (absorbance 600-650 nm)

Calculating phosphate

Based on your standard curve, the equation is gotten as below:

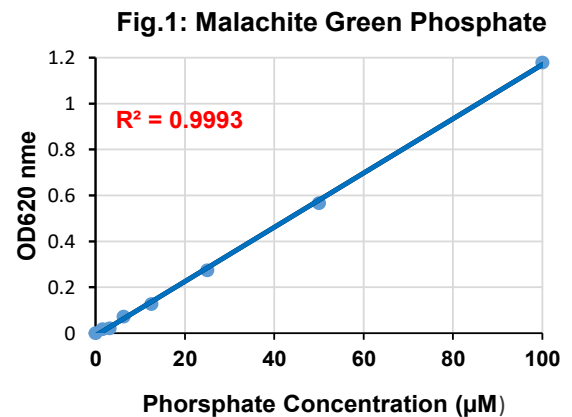
Equation 1: $Y=A*X + B$

The phosphate amount X (μM) = (Y-B)*DF/A

Y= Sample OD; A= Slope;

B=constant value; DF=dilution factor.

The phosphate standard curve is shown in Fig. 1.



RELATIVE PRODUCTS

- CD38 Cyclase Activity Assay (TBS2100)
- L-Lactate colorimetric assay (TBS2071)
- LDH Cytotoxicity Assay (TBS2002)
- Resazurin Cell Viability (TBS2001)
- CCK-8 Cell Viability Assay (TBS2022)
- Homocysteine Fluorometric Assay (TBS2091)
- AHCY Inhibitor Screening Assay (TBS2097)
- G6PDH Activity Colorimetric Assay (TBS2102)
- ATP Colorimetric/Fluorometric Assay (TBS2010)
- ADP Colorimetric / Fluorometric Assay (TBS2020)
- Caspase-3 Colorimetric Assay (TBS2030)
- NNMT Inhibitor Screening Assay (TBS2097)
- NNMT Activity Assay (TBS2098)
- Caspase-3 Colorimetric Assay kit (TBS2030)
- Mitochondria Isolation (TBS2116)
- Mitochondria Complex 1 Activity Assay (TBS2017)
- Mitochondria Oxidase Activity (TBS2105)
- Mitochondrial Membrane Potential Assay (TBS2049)
- Cytochrome C Oxidase Activity Assay (TBS2115)

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