Tribioscience

Glucose 6-Phosphate Activity Colorimetric Assay

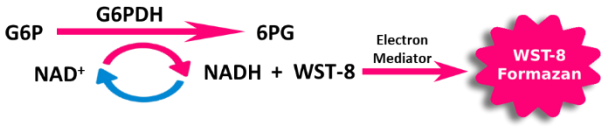
(TBS2103, 100 Assays, Store at -20°C)

DESCRIPTION

Glucose-6-phosphate (G6P) is a key intermediate in the metabolism of glucose, and its levels can be indicative of various physiological processes. G6P is formed from glucose phosphorylation by the enzyme hexokinase. Further, it is oxidized by G6P dehydrogenase (G6PD) in present $NAD(P)^+$ to 6-Phosphogluconolactone (6PG) and NAD(P)H.

Tribioscience's G6P Colorimetric Assay kit provides a flexible, accurate, sensitive, and time-saving approach for detecting G6P in a wide variety of samples. In this assay, G6PD converts G6P and $NAD(P)^+$ to 6PG and NAD(P)H, which in turn induces the Colorimetric Probe to generate intense colored product at 460 nm as shown in Fig.1.

Fig.1. Principle of the Assay.



FEATURES

- **Flexible:** Suitable for 96-well or 384-well plate.
- **Accuracy**: Absorbance measurement is proportional to the G6P concentration.
- **Sensitive:** Can detect as low as 1.0 nm/well.
- **Time-saving**: Just incubate and read out style.

Kit Components for 100 Assays, store at -20°C.

Part Name	Part Size (100 tests)
G6P Substrate Stock	20 µL
G6P Colorimetric Probe	1.2 mL
G6P Assay Buffer	10 mL
G6PD Enzymes	30 µL
G6P Standard Stock (2 mM)	200 µL

APPLICATIONS

- Measure G6P concentration in a variety samples like cells and tissues.

DIRECTIONS FOR USE

Sample and Enzyme Preparation

1. Cells or Tissues are homogenized in 3 volumes of the assay Buffer.
2. Centrifuge the samples at 2000xg for 10 min at 4°C.
3. Take the supernatant to new tube on the ice. If not do the assay the same day, freeze the samples at -80°C for a month.

Assay Procedures

1. P6G Standard Preparation as Table below.

Tube #	G6P standard (µL)	Assay Buffer (µL)	P6G (µM)	P6G (nmol/well)
1	56.3	93.8	750	37.50
2	37.5	112.5	500	25.00
3	18.8	131.3	250	12.50
4	7.5	142.5	100	5.00
5	3.8	146.3	50	2.50
6	1.9	148.1	25	1.25
7	0	150	0	0.0

2. Load samples:
Add 50 µL of Standard, or sample, or blank control to indicated each well. (Note: recommend running a pilot study to determine the optimal concentration of sample within the assay standard curve range).

3. Assay Working Reagent Preparation for 100 tests: please adjust the volume as you need.
Mixing the below reagent for 100 tests: total volume is 5 mL.

- 3990 µL Assay Buffer.
- 20 µL Enzyme solution.
- 10 µL Substrate Stock solution
- 1000 µL Detection Probe.

4. Add 50µL of above assay working reagent to each well containing the standards, diluted positive control, and the test samples.

5. Incubate at room temperature for about 20 ~ 60 minutes, with gentle shaking and protected from light.

6. Measure $OD_{460\ nm}$

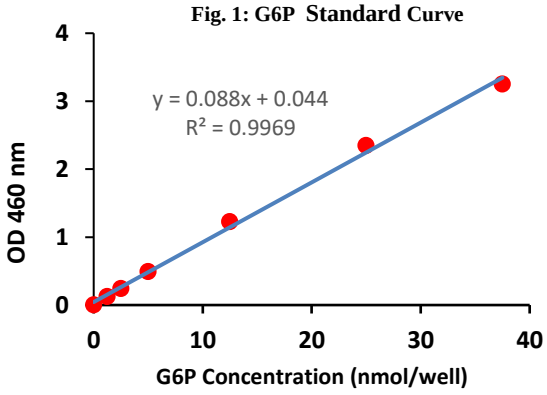
7. Concentration Calculation:

Calculate $\Delta A\ (OD_{460\ nm}) = A_{sample} - A_{blank}$

G6P concentration (µM) = DF* ΔA/slope

DF= Dilution Factor

Fig. 1: G6P Standard Curve



RELATED PRODUCTS

6-Phosphogluconate Colorimetric Assay (TBS2089C)
Glucose-6-phosphate dehydrogenase Activity Colorimetric Assay (TBS2102C)
ATP Colorimetric/ Fluorometric Assay (TBS 2010)
ADP Colorimetric/Fluorometric Assay (TBS3020)
Hexokinase Activity Colorimetric Assay (TBS2107C)
6-Phosphogluconate Dehydrogenase Activity Colorimetric Assay (TBS2093C)
Resazurin Cell Viability (TBS2001)
LDH Cytotoxicity Assay (TBS2002)
Catalase Assay (TBS2006)
CCK-8 Cell Viability Assay (TBS2022)
Caspase-3 Colorimetric Assay (TBS2030)
Cytochrome c Reductase Activity Assay (TBS2116)
AOPI Viability Assay for Flow Cytometry (TBS2069)

For research use only