Glutamate Quantification Colorimetric Assay (Catalog: TBS2014, 100 Assays, Store at -20 ℃)

DESCRIPTION

Glutamate, also known as L-glutamic acid, is the most abundant excitatory neurotransmitter in the brain. It plays a crucial role in learning and memory. Dysfunction of glutamate in the brain is associated with many brain diseases, such as Parkinson's disease, Alzheimer's disease and Huntington's disease.

Tribio™ Glutamate Quantification Colorimetric Assay is based on a dehydrogenase coupled reaction converting chromophore to a colored formazan, which can be measured at OD 460 nm. The generated signal is proportional to the content of glutamate in the samples. The kit provides the easiest and most accurate approach to measuring glutamate from a variety of samples.

Synonyms: Glutamic acid; Glu; 2-Aminoglutaric acid.

APPLICATIONS

Specifically detect glutamate from cell and biological tissues.

KIT CONTENTS FOR 100 TESTS:

Name	Part Size
Assay Buffer	10 mL
Enzymes	0.4 mL
Enzyme substrate (20x)	0.3 mL
Glutamate Detection Probe (5x)	1.2 mL
Standard stock (5x)	0.3 mL

Storage conditions: Store the Reagent at –20 ℃ for a year.

PROCEDURES

1. Sample preparation

1) Wash the cells with 400 µL cold PBS.
2) Pellet 2 X 105 cells at 2000 rpm for 5 min.
3) Aspirate the wash solution from the tube.
4) Homogenization in 150 µL assay buffer by Vortex for 30 sec.
   (For tissues, weigh ~20 mg tissue & wash with cold PBS. Homogenize in Assay Buffer in a micro-centrifuge tube).
5) Centrifuge at 14000 rpm for 5 min.
6) Transfer the extracted supernatant into 2 labeled tubs.
7) To transfer 50 µL of extracted samples into labeled 96-well plate.

2. Prepare premix for 50 µl/well premix

2.5 µL of enzyme substrate + 3 µL of enzyme + 10 µL of glutamate detection probe + 34.5 µL assay buffer.

3. Prepare standards as below Table 1.

Table 1: Glutamate Standard Preparation

Std	Standard (µL)	Assay buffer (µL)	µM	nmol/well
1	100 (10 mM stock)	400	2000	100
2	150 (Std1)	150	1000	50
3	150 (Std1)	150	500	25
4	150 (Std1)	150	250	12.5
5	150 (Std1)	150	125	6.25
6	150 (Std1)	150	62.5	3.125
7	150 (Std1)	150	31.25	1.563
8	0	150	0	0

4. Set up reaction:

1) Add 50 µL of Standard or test sample preparation, or blank (assay buffer) into the indicated well. (Note: recommend running a pilot study to determine the optimal concentration of sample within the assay standard curve range).
2) Add 50 µL of premix to each well containing the standards, test samples, and blank.

5. Initiate Reaction:

Incubate at 37°C for about 30 minutes, with gentle shaking and protecting from light.

6. Measure OD value at 460 nm:

The glutamate standard and samples can be read at the end point.

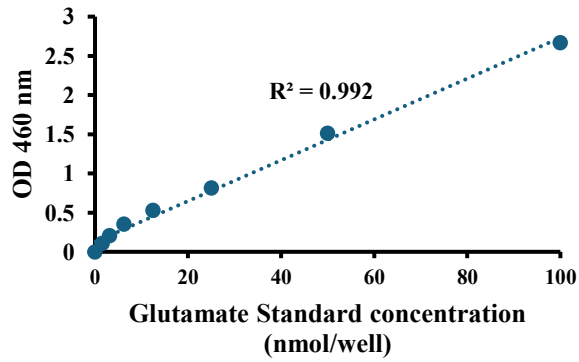
7. Calculation using standard curve:

Apply the sample OD reading to the standard curve to get the total glutamate amount in the sample wells.

TYPICAL STANDARD CURVE

The Typical Standard curve showed as an example in Fig.1. It can be used only for reference.

Fig. 1: Glutamate standard curve



RELATIVE PRODUCTS

Resazurin Cell Viability Kit (TBS2001)
LDH Cytotoxicity Assay (TBS2002)
Caspase-3 Colorimetric Assay kit (TBS2030)
CD38 Cyclase Activity Assay (TBS2100)
LDH Activity Assay (TBS2012)
CCK-8 Cell Viability Assay (TBS2022)
GOT Activity Assay (TBS2013)
Thiol Fluorometric Assay (TBS2026)
GSH Assay (TBS2028)
Homocysteine Fluorometric Assay (TBS2091)
AHCY Inhibitor Screening Assay (TBS2097)
G6PDH Activity Colorimetric Assay (TBS2102)
ATP Colorimetric/Fluorometric Assay (TBS2010)
ADP Colorimetric / Fluorometric Assay (TBS2020)
NNMT Activity Assay (TBS2098)
NADP Assay (TBS2058)
NAD/NADH Assay (TBS2029)

For research use only.