# **PractiChrom<sup>™</sup> L-Glutamate Assay Kit (PGLT-25)**

Quantitative L-Glutamate Determination Using PICOEXPLORER<sup>™</sup>

## DESCRIPTION

Glutamate is an important chemical in general metabolism. It is also widely used as a flavor enhancer in the food industry. Consumption of foods containing monosodium glutamate (MSG) has been known to cause headaches, flushing, sweating, nausea, and more. Glutamate is also a crucial mammalian neurotransmitter that is believed to be involved in a number of neurological and psychiatric disorders such as lateral sclerosis, autism, and Alzheimer's disease.

BioAssay Systems' glutamate assay kit is based on glutamate dehydrogenase catalyzed oxidation of glutamate in which the formed NADH reduces a chromogenic reagent. The intensity of the product color is directly proportional to the glutamate concentration in the sample.

## **KEY FEATURES**

**Sensitive and accurate**. Detection limit of 0.05 mM (7.3 ppm, 0.73 mg/dL) and linearity to 2 mM (292 ppm, 29.2 mg/dL) Glutamate.

Convenient. Assay performed with portable PiCO Explorer device.

Cost efficient. No need for expensive plate readers.

## APPLICATIONS

**Direct Assays:** Glutamate in food and beverage samples (e.g. soups, sauces, milk, etc).

## **KIT CONTENTS (25 TESTS)**

Assay Buffer:	5 mL	Standard:	1.0 mL 100 mM Glutamate
Enzyme A:	30 µL	NAD/MTT:	1.0 mL
Enzyme B:	120 µL		

**Storage conditions**: The kit is shipped on ice. Store all components at -20°C upon receiving. Shelf life: 6 months after receipt.

**Precautions**: Reagents are for research use only. Equilibrate all components to room temperature prior to assay. Normal precautions for laboratory reagents should be exercised while using the reagents. Please refer to Material Safety Data Sheet for detailed information.

## PROCEDURES

#### Sample Preparation:

Fish Sauce & Soy Sauce. Dilute 200× in dH\_2O by adding 5  $\mu L$  sample to 995  $\mu L$  dH\_2O.

Soups. Dilute 40× in dH<sub>2</sub>O by adding 5  $\mu$ L sample to 195  $\mu$ L dH<sub>2</sub>O.

*Milk* samples should be cleared by mixing 600  $\mu$ L milk with 100  $\mu$ L 6 M HCl. Centrifuge 5 min at 14,000 rpm. Transfer 300  $\mu$ L supernatant into a clean tube and neutralize with 50  $\mu$ L 6 M NaOH. The neutralized supernatant is ready for assay (dilution factor n = 1.36).

Samples that are known to contain high levels of glutamate or samples that are colored will require a dilution. For preparation protocols for samples not listed above, please contact our technical support at info@bioassaysys.com

#### Procedure

- 1. Prepare 2 mM Glutamate Standard by mixing 5  $\mu L$  of the provided 100 mM Standard and 245  $\mu L$  dH\_2O in an Eppendorf tube.
- 2. In separate PCR tubes, add 10  $\mu L$  dH\_2O and 10  $\mu L$  2 mM Glutamate Standard.

Samples. Add 10 µL Sample to a PCR tube.

Reagent Preparation. Prepare sufficient Working Reagent for all dH<sub>2</sub>O, Standard, and Sample tubes by mixing, for each tube: 40  $\mu$ L Assay Buffer, 4  $\mu$ L NAD/MTT, 0.5  $\mu$ L Enzyme A, and 0.5  $\mu$ L Enzyme B.

Then quickly add 40  $\mu$ L Working Reagent to all dH<sub>2</sub>O, Standard, and Sample tubes. Close the tubes, briefly vortex or tap to mix. Tap tube on bench to settle liquid to the bottom of the tube if needed. Incubate for 10 min at room temperature in the dark.

3. Please refer to the PICOEXPLORER<sup>™</sup> User's Manual for detailed instructions for operating the device.

Download the PAS-110 application. Turn on Bluetooth.

Push the Power button on the device. Then, open the app and tap the Connection Setting button and connect the device.

## Measuring a Standard Curve (See pg 17-19 in User's Manual)

Return to the main menu and tap the Standard Curve button. Set the following:

LED Output: 10%

Unit: mM

**RBG Selection: G** 

Tap the first Known Concentration Data Input Area box and input 0.0. Then, tap on the second box and input 2.0 (this represents the 0 and 2 mM Glutamate Standards). Then, place the dH<sub>2</sub>O tube into the measurement chamber of the photo absorbance sensor. Tap the Known Concentration Measurement Input Area (the box below 0.0), and click Measure. Remove the tube, then place the 2 mM Standard into the measurement chamber. Tap the box below 2.0 and click Measure. Click Graph to view the standard curve.

#### Measuring Sample Concentrations

Return to the main menu and tap the Measure button. Edit the LED output, Units, and RBG selection as done above for the standard curve.

Place each Sample into the measurement chamber of the photo absorbance sensor and tap Measure.

## CALCULATION

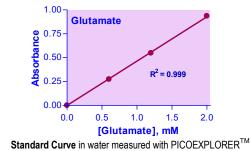
The "concentration" will be displayed on the PICOEXPLORER<sup>TM</sup> for each Sample. To calculate the glutamate concentration in the sample, multiply the Sample concentration by the dilution factor used (e.g. 200, 40, 2, 1.36, etc).

[Glutamate] = [Sample] × n (mM)

where [Sample] is the concentration of sample and *n* is the dilution factor. Note: if the sample concentration says "Out of range" the sample is not within the linear range of the assay. If the color of the tube is yellow like the dH<sub>2</sub>O tube, then the sample has low levels of glutamate that cannot be detected by the assay. If the sample is very dark, dilute further in water and repeat the assay. Multiply the results by the dilution factor. **Conversions**: 1 mM glutamate 14.6 mg/dL, or 146 ppm.

## MATERIALS REQUIRED, BUT NOT PROVIDED

Pipetting devices, PCR tubes (e.g. Watson 137-211c 0.2 mL; or Cat# PCR-50 from BioAssay Systems), Eppendorf tubes (e.g. Phenix Cat# MAX-715, or Cat # EPP-50 from BioAssay Systems), and PICOEXPLORER<sup>TM</sup> (Cat # PICO001).



## **RELATED PRODUCTS**

PICOEXPLORER<sup>™</sup> (Handheld Colorimeter): Cat # PICO001. PCR Tubes: Cat # PCR-50, Pack of 50 tubes Eppendorf tubes: Cat # EPP-50, Pack of 50 tubes L-Glutamate Quick Test Strips: Cat # QQGLUT10.