Monoamine Oxidase (MAO) Assay Kit(Colorimetric)

Catalog No.: K059

Size:50T/100T

Storage: The kit should be stored at 2-8℃ for one year.

Intended use:

This kit can be used to measure monoamine oxidase (MAO) activity in serum, plasma and animal tissue samples.

Sensitivity: 16 U/L

Detection range: 16-641 U/L

Introduction

MAO can catalysis 4-dimethylambenzylamine to produce p-dimethylaminobenzaldehyde. p-Dimethylaminobenzaldehyde has a characteristic absorption peak at 355nm. The activity of MAO can be calculated indirectly by analyzing the production of p-dimethylaminobenzaldehyde.

Kit Components

Item	50T	100T
Reagent 1	30mL	60mL
Reagent 2	60mL	60mL*2
Reagent 3	60mL	60mL*2
Reagent 4	3mL	5mL

Materials prepared by users

Microplate Reader(355 nm)

Reagent preparation

1.The preparation of Reagent 1 working solution:

Mix Reagent 1 with double distilled water fully at a ratio of 1:1. The prepared solution can be stored at 2-8℃ for 1 month.

2.The preparation of Reagent 3 working solution:

Mix reagent 3 with double distilled water fully at a ratio of 1:1.The prepared solution can be stored at

2-8℃ for 1 month.

Sample preparation

1.Serum or plasma: detect directly.

2.10% tissue homogenate: Accurately weigh the tissue 0.1-0.5g, then add 9 times the volume of pre-cooled Reagent 1 working solution according to the ratio of Weight (g): Volume (mL)=1:9. The sample is mechanically homogenized for 90s in an ice water bath. Centrifuge at 1000 g for 10 min at 4°C, then take the supernatant(determine the protein concentration of supernatant (K001) before centrifugation) and centrifuge at 10000 g at 4°C for 30 min, discard the supernatant and retain the precipitation. Add 1 mL of pre-cooled Reagent 2 and mix fully, centrifuge at 16,000 g at 4°C for 40 min, discard the supematant and retain the precipitation. Finally, add 1 mL of pre-cooled Reagent 3 working solution, mix fully, and store it on ice for detection.

Note: Reagent 1 working solution, Reagent 2, Reagent 3 working solution need to be pre-cooled for 30min in advance.

Assay Procedure

1. Reagent 3 working solution and Reagent 4 need to be pre-heated at 37° C for 30 min in advance.
2. Add reagents according to the table below:

reagents	Sample tube(μ L)
Sample	25
Reagent 3 working solution	150
Reagent 4	25

3. Vibrate on Microplate Readerr for five seconds, measure the OD value of samples at 355 nm, recorded as A_1, and then incubate accurately at 37℃ for 30 min, measure the OD values of each samples again, recorded as A_2.

Calculation

1. Serum or plasma sample:

Definition: the amount of enzyme in 1 L of serum (plasma) that catalyze the substrate to produce 1 nmol p-dimethylaminobenzaldehyde at 37 C for 1 min is defined as 1 unit.

$$\text{MPO activity(U/L)} = \frac{A_2-A_1}{\varepsilon \times d} \times V_1 \div V_2 \div T$$

2. Tissue sample:

Definition: the amount of enzyme in 1 g of tissue protein that catalyze the substrate to produce 1 nmol p-dimethylaminobenzaldehyde at 37℃ for 1 min is defined as 1 unit.

$$\text{MPO activity(U/gprot)} = \frac{A_2-A_1}{\varepsilon \times d} \times V_1 \div (V_2 \times C_{pr}) \div T$$

Note:

T: the time of incubation in the reaction, 30min

ε : the molar extinction coefficient of p-dimethylaminobenzaldehyde, 2.77×10-4L/nmol.cm)

d: the optical path of cuvette, 0.6cm.

V_1: the total volume of reaction, 200 μl.

V_2: the volume of sample, 25 μl.

C_{pr}: The concentration of protein in sample, gprot/L.