

Tyrosine Colorimetric Assay (TBS2070, 100 Assays, Store at -20°C)

Description

Tyrosine (Tyr) is an aromatic amino acid derived from phenylalanine. It serves as a precursor for thyroid hormones, catecholamines, neurotransmitters, and melanin, including epinephrine, norepinephrine, dopamine, thyroid hormones, melanin, fumarate, and acetoacetate. Its metabolism is involved in energy production. The pathology of abnormal concentrations of Tyr is well-known in diseases including phenylketonuria, hypothyroidism, tyrosinemia, albinism, alkaptonuria and neurodegenerative diseases.

Tribioscience's Tyrosine Colorimetric Assay kit is a simple, and sensitive approach that can detect normal and abnormal concentrations of Tyr in biological fluids. The assay is based on the enzymatic oxidation of Tyr producing a stable signal (OD 410 nm), which is directly proportional to the amount of Tyr. Sample preparation is simple and does not require strenuous or complicated procedures. The assay can detect as low as 8 µM of Tyr in a variety of biological samples.

Synonyms: Tyr, L-Tyr, 4-Hydroxyphenylalanine, S-tyrosine, and p-tyrosine, L-Tyrosine.

Assay Principle

Enzyme **Probe**
Tyrosine → **Intermediate** → **Chromophore (OD 410 nm)**

Application

- Measurement of Tyrosine in various biological samples.
- Analysis of Tyrosine in pathological conditions.

Kit Contents

Component	100x Rxns
Tyrosine Enzyme	130 µL
Tyrosine Probe	0.6 mL
Tyrosine Standard (100 mM)	20 µL
Assay buffer	13 mL

Storage conditions: Store the kit at -20°C protected from light.
Shelf life: 12 months.

Assay Procedures

1. Sample Preparation

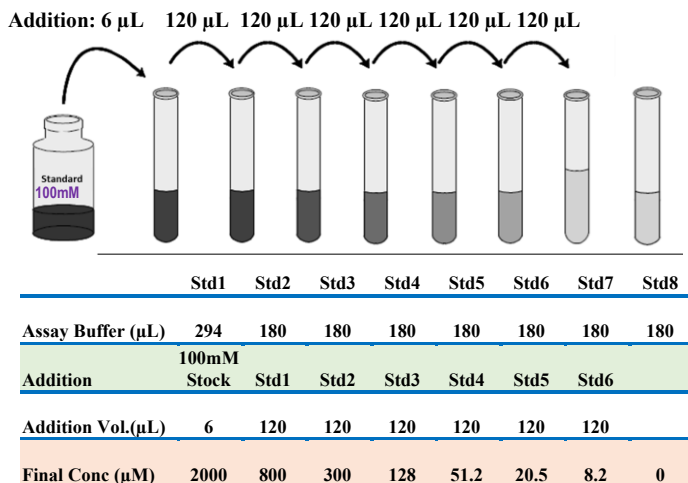
Collect cell culture supernatant, serum, plasma, urine, and other biological fluids. Deproteinizing the samples using 10 kDa Spin Column (Amicon Ultra 0.5 mL, Cat. No. UFC501024). Briefly, add sample to the spin column, centrifuge at 10,000 x g for 5 min. at 4°C. Collect the filtrate. Add 50 µl of filtrate into desired well(s) in 96-well plate.

2. Standard Preparation as Fig.1:

2.1 Label 8x 1.5mL tubes 1-8 for a standard curve as shown in the Diagram (Fig. 1).

- 2.2 Add 294 µL of Assay Buffer to Std 1 and 180µL to Std 2-8.
- 2.3 Add 6 µL of 100 mM tyrosine Standard Stock solution to Std1, then carry out a 2.5 x serial dilution through Std 2-7. Leave Std8 as the 0 standard (the assay buffer alone). The standards concentrations are 2000, 800, 320, 128, 51.2, 20.5, 8.2 and 0 µM from Std1 to Std0 respectively.

Fig.1 Diagram for Tyrosine Standard Preparation



- 2.4 Load the samples: Pipet 50µL of standards, controls, and test samples into individual wells of a microplate in duplicate manner (*Note: recommend running a pilot study to determine the optimal concentration of sample within the assay standard curve range*).
- 2.5 Prepare Enzyme working solution for each well by mixing the reagent as follows:
- 43.8 µL Assay solution
 - 1.2 µL Enzyme
 - 5 µL probe
- 2.6 Begin the reactions. Add 50µL of above working solution to each microplate well containing the standards, controls, and samples.
- 2.7 Incubate the reactions. Incubate at 37°C for 90-120 minutes protected from light.
- 2.8 Measure absorbance using a microplate reader with endpoint at 410 nm.
- 2.9 Correct for background absorbance. For each point, subtract the value derived from the 0 control.

Calculation

Subtract the blank value (0µM Standard) from the standard values and plot the ΔOD against standard concentrations. Determine the slope and calculate the Tyrosine concentration of the Sample using the equation obtained from the linear regression of the standard curve.

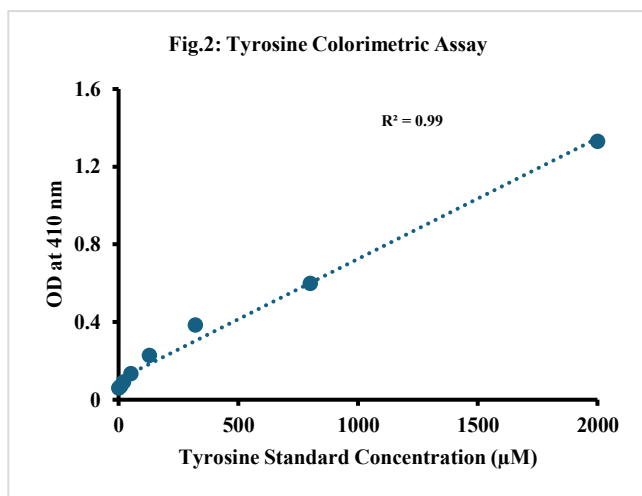
$$\text{Ty Concentration (µM)} = N \times (\text{Rsample} - \text{Rblank}) / \text{Slope (µM)}$$

Where: Rsample and Rblank are optical density readings of the sample and blank, respectively. N is the sample dilution factor.

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Typical Data

This standard curve is provided for demonstration only as below **Fig.2**. A standard curve should be generated for each set of samples assayed.



Related Products

Tyrosinase Activity Assay (TBS2072)
 ATP Activity Assay (TBS2010)
 Hydrogen Peroxidase Activity Assay Kit (TBS2067)
 HRP Fluorescence -System (TBS5026)
 Tryptase Activity Assay (TBS2101)
 β-Hexosaminidase Activity Assay (TBS2105)
 Cytochrome C Oxidase Activity Assay (TBS2115)
 Fast Glucose Determination Colorimetric/Fluorometric Assay (TBS2087)
 Glucose Oxidase Activity Colorimetric/Fluorometric Assay (TBS2088)
 Non-esterified Fatty Acid Assay (TBS2203)
 Glycerol Colorimetric / Fluorometric Assay (TBS2204)
 Protein Assay Kits (TBS2005)
 Cell Nuclear Extract kit (TBS6025)

For research use only.