

(FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS!)

β -glucuronidase (Exogenous) Assay Kit

Catalog No.: BC00219

Size: 100T

Please read the instructions carefully before use. If you have any questions or need further help during experiment, please don't hesitate to contact us through the following methods:

✉ Email (Sale)	order@enkilife.com
✉ Email (Techsupport)	techsupport@enkilife.com
☎ Tel:	0086-27-87002838
🌐 Website:	www.enkilife.com

Shelf life: Please refer to the label on the outer package.

Techsupport: In order to provide you with better service, please inform us the lot number on the label of the outer package.

Basic Information

Product Name	β -glucuronidase (Exogenous) Assay Kit
Detection Method	Colorimetric
Sample Type	Tissue
Assay Type	Quantitative
Detection Instrument	Microplate reader (540 nm)

Product Introduction

β -Glucuronidase (β -GD) is widely present in animal tissues. As a matrix-degrading enzyme involved in tumor invasion and metastasis, it physiologically hydrolyzes steroid glucuronides and acid mucopolysaccharides. This enzyme is abundant in hepatocytes as well as gastric cancer tissues. The detection of β -GD activity in gastric juice is of great significance for gastric cancer research.

Principle

β -Glucuronidase acts on the specific substrate to release free phenolphthalein. The enzyme activity can be determined by colorimetry based on the quantity of free phenolphthalein.

Components

No.	Components	Size	Notes
Reagent 1	Powder	1 vial	Store at below -20°C.
Before use, add 1 mL of Reagent 2 to each vial of powder to prepare Working Solution 1, mix thoroughly, and store at below -20°C.			
Reagent 2	Liquid	6 mL	Store at 4°C.
Reagent 3	Liquid	40 mL	Store at 4°C.
Reagent 4	Standard powder	1 vial	Store at 4°C and protect from light.

	Standard solvent	5 mL	Store at 4°C.
Before use, dissolve one vial of standard powder with 1 mL of standard solvent to prepare 10 µmol/mL phenolphthalein standard solution and mix thoroughly. Store the prepared solution at 4°C. Then dilute the 10 µmol/mL standard solution 5-fold (1:4) with standard solvent to obtain 2 µmol/mL phenolphthalein standard solution. Prepare freshly before use.			
Consumable 1	Microplate	1 plate	Store at room temperature.
Consumable 2	Plate Sealer	2 pieces	Store at room temperature.

Storage

The kit can be stored for 3 months under specified conditions.

Preparation

Equipment and reagents required: Microplate reader (wavelength: 540 nm), 96-well plate, 37 °C water bath or incubator, tabletop centrifuge, pipettes of various specifications, double distilled water, 0.9% normal saline or 0.1M PBS, vortex mixer, test tubes or centrifuge tubes, protein assay reagents (available from our company).

• Sample requirements

Tissue: Accurately weigh the tissue sample. Add 9 volumes of normal saline at a ratio of tissue weight (g) to volume (mL) = 1:9, then homogenize under low temperature (0-4 °C). Centrifuge at 4000 rpm for 10 minutes, and collect the supernatant for testing.

Operation process

Can be performed in 0.5 mL or 1.5 mL centrifuge tubes.

	Blank well	Standard well	Assay well
Double distilled water (μL)	10		
2 μmol/mL phenolphthalein standard solution (μL)		10	
Supernatant (μL)			10
Reagent 1 Working Solution (μL)	10	10	10
Reagent 2 (μL)	40	40	40
Mix well and incubate at 37 °C for 1 hour.			
Reagent 3 (μL)	400	400	400
Mix thoroughly, then centrifuge at 8000 rpm for 10 minutes. Transfer 200 μL of the supernatant from each tube to a 96-well plate, and measure the absorbance (A) of each well using a microplate reader at 540 nm.			

Calculation

1. Calculation for liquid samples such as serum

Definition: One unit (U) of enzyme activity is defined as the amount of enzyme that catalyzes the production of 1 μmol phenolphthalein per minute per milligram of tissue protein at 37°C.

$$\beta - \text{Glucuronidase Activity (U/mg)} = \frac{A_{\text{Assay}} - A_{\text{Blank}}}{A_{\text{Standard}} - A_{\text{Blank}}} \times C_{\text{Standard}} \div T \div C_{\text{pr}}$$

Annotation:

C_{Standard} : Standard solution concentration, 2 μmol/mL

C_{pr} : Tissue homogenate protein concentration, mg/mL

T: Reaction time, 60 min

Precautions

1. It is recommended to use disposable laboratory test tubes for operation.
2. This product is intended for scientific research use only by professionals and must not be used for clinical diagnosis or treatment, in food or drugs, or stored in ordinary residences.
3. Prepare reagents strictly in accordance with the instructions and protect from light.
4. Conduct pre-experiments for tissue homogenate in advance to determine the optimal sampling concentration.