Lipase Assay Kit

(No.:BC003 Size:96T Microplate method))

1. Packaging specification

Reagent 1: 18 mL × 1 bottle
Reagent 2: 6 mL × 1 bottle
Standard: 200 µL × 1 vial (concentration indicated on the label)

2. Intended use

For the determination of lipase activity in serum, plasma, tissue homogenates, cells (or cell supernatants).

3. Assay principle (Microplate method)

1,2-O-dilauroyl-rac-glycerol-3-pentanoate-(6-methylresorufin) ester + H_2O → 1,2-O-dilauroyl-rac-glycerol + pentanoate-(6-methylresorufin) ester,
pentanoate-(6-methylresorufin) ester → pentanoic acid + 6-methylresorufin (color development).
At a wavelength of 580 nm, lipase activity is determined according to the generation rate of the red-colored product methylresorufin.

4. Storage conditions and validity

Reagents are stable for 6 months when stored at 2–8 ℃ protected from light. Do not freeze. Do not transport at high temperature. After opening, store at 2–8 ℃ protected from light and stable for 1 month; it is recommended to use as soon as possible within 7 days.

5. Applicable instruments

All types of microplate readers (580 nm wavelength).

6. Sample requirements

Blood: Serum or plasma should be separated promptly after collection, avoiding hemolysis. Serum (plasma) enzyme activity is stable for 1 day at 2–8 ℃ and for 1 month at −20 ℃. Thawed samples can be used only once.
Tissue samples: Accurately weigh the tissue, add 9 volumes of normal saline according to the ratio of weight (g): volume (mL) = 1:9, mechanically homogenize in an ice-water bath to prepare a 10% homogenate, centrifuge at 2500 rpm for 10 minutes, and collect the supernatant for determination.
Cell samples should be collected, disrupted, and prepared into homogenates for determination; **cell culture supernatants** can be directly used for the assay.
***Note:** Tissue or cell samples must be tested on the same day after homogenization.

7. Assay method

1、Main performance parameters:

Wavelength	580nm	Reaction temperature	37℃	Reaction method	Rate method
Calibration method	Two-point calibration	Calibration type	Linear	Reaction direction	Upward

2、Operation procedure

Reagent \ Well	**Blank**	**Standard**	**Sample**
Double distilled water (μL)	4		
standard solution (μL)		4	
Sample (μL)			4
Reagent 1 (μL)	180	180	180
Mix well, incubate at 37°C for 3-5 minutes			
Reagent 2 (μL)	60	60	60
Mix well, incubate at 37 ℃ for 1 minute, read A0 at 580 nm on the microplate reader, then read A2 again after 2 minutes, calculate △A=A_2-A_0			

3. Calculation formula:

$$\text{Serum/Plasma LPS Activity(U/L)} = \frac{\Delta A_{sample} \quad \Delta A_{blank}}{\Delta A_{Calibrator} \quad \Delta A_{blank}} \times C_{Calibrator}$$

$$\text{Tissue / Cell } LPSActivity\text{（U/gprot）} = \frac{\Delta A_{sample} \quad \Delta A_{blank}}{\Delta A_{calibrator} \quad \Delta A_{blank}} \times C_{standard} \div Cpr$$

$C_{calibrator}$: concentration of standard solution, U/L;

Cpr: protein concentration of homogenate, gprot/L (prot refers to protein).

8. Reference range

Human serum (plasma): 1–60 U/L. This reference range is for reference only; it is recommended that each laboratory establish its own reference range.

9. Product performance indicators

Reagent blank absorbance: A_{580nm} (1.0 cm) ≤ 0.300;

Linear range: 15–300 U/L (criterion: r^2 ≥ 0.990);

Accuracy: relative deviation ≤ 10.0%; intra-assay CV ≤ 6.0%; inter-assay relative range ≤ 10.0%

10. Precautions

1. EDTA, oxalate, fluoride, and sodium citrate have inhibitory effects on enzyme activity.
2. Conjugated bilirubin ≤ 20 mg/dL, ascorbic acid ≤ 20 mg/dL, free bilirubin ≤ 20 mg/dL have no effect on the results.
3. If the sample concentration exceeds the linear range, dilute the specimen with normal saline; multiply the result by the dilution factor.
4. If the required wavelength is not available on the instrument, select a nearby wavelength (±10 nm).
5. Mixing reagents from different batches is not recommended.

6. Some raw materials in this kit are derived from animals and microorganisms; please take protective measures during use and strictly follow laboratory operating procedures. Waste liquids should be disposed of according to environmental protection requirements.
7. If LPS activity in the sample is low, the second reading time may be extended (e.g., 5 minutes or 10 minutes).
8. This kit is for laboratory and research use only. Please read the instructions carefully before use.