Glucose (GLU) Test Kit

(Cat/No.: BC036   Size:96T)

1.Compositions & Preparation(The kit is valid for 1 year)

Reagebt	Specification	Component	Concentration	storage
Working fluid	25mL×1 bottle	**Phosphate buffer**	100mmol/L	Store at 2～8℃ avoid light for 1year
		DHBS	2.0mmol/L	
		4-Aminoantipyrine	1.0mmol/L	
		glucose oxidase	10kU/L	
		Magnesium chloride	3.5mmol/L	
		peroxidase	8kU/L	
Standard	0.1mL×1 stick	glucose	Concentration see label	
It comes with a disposable 96-well plate				

2. Assay Pricinple

$$\text{Glucose}+H_2O+O_2 \xrightarrow{\text{GOD}} \text{D-Glucise}+H_2O_2$$

$$H_2O_2+\text{4 - aminoantipyrine + DPBS} \xrightarrow{\text{POD}} \text{redpurple pigment}$$

Tested at 505nm, its color is proportional to the glucose concentration.

3. Operation process

1).Sample preparation

1 、Serum (or heparin anticoagulated plasma): direct determination, such as determination after dilution with normal saline beyond the linear range.

②、Culture medium sample: aspirate the culture medium, centrifuge at 1000 rpm for 10 minutes, and take the supernatant for determination. [Note]: It is generally recommended that the cell density be above 1 million cells/mL.

③Tissue sample: Accurately weigh the tissue, according to the ratio of weight (g): volume (mL) = 1:9, add 9 times the volume of normal saline (or PBS), mechanically homogenize under ice-water bath conditions, 2500 rpm , centrifuge for 10 minutes, and take the supernatant for testing.

④Cell samples: A. Cell collection: Take out the prepared cell suspension, centrifuge at 1000 rpm for 10 minutes, discard the supernatant, and keep the Cell pellet; wash with isotonic buffer (recommended 0.1mol/L, pH7-7.4 phosphate buffer) for 1-2 times, the same 1000 rpm, centrifugation for 10 minutes, discard the supernatant, and leave the cell pellet; B. Cell disruption: add 0.2-0.3mL of homogenization medium (recommended 0.1mol/L, pH7-7.4 phosphate buffer or physiological saline) for homogenization, and ultrasonically disrupt under ice-water bath conditions (power 300W, 3-5 seconds) /time, with an interval of 30 seconds, repeat 3 to 5 times) or manual homogenization, the prepared homogenate is measured directly without centrifugation. Lysis solution can also be used for lysis (TritonX- 100 is recommended, 1-2%, lysis time is 30-40 minutes), and the lysed liquid can be measured directly without centrifugation. [Note]: It is generally recommended to collect cells at a density of more than 1 million cells/mL. The broken liquid can be observed by microscope to see if the cells are completely broken.

2) . Operation table

96-well plate operation, microplate reader colorimetric			
	Blank	stadnard	sample
Distilled water	2.5		
Starndard (μL)		2.5	

sample（μL）			2.5
Working fluid （μL）	250	250	250

Gently shake the well plate, incubate at 37°C for 10 minutes, wavelength 505nm, and measure the absorbance value of each well with a microplate reader.

Fully automatic machine operation			
Sample size/water/standard	Sample Volume	μL	2.5
Working fluid	reagent	μL	250
Incubate at 37°C for 10 minutes, adjust the working solution + distilled water to zero, and measure the light absorption value A.			
	Main wavelength	nm	505
type of reaction	Reaction type		endpoint method
Reaction direction	Reaction direction		L reaction (+)

4. Calculation formula and example

1). Calculation formula for liquid samples such as serum:

Microplate reader clorimetric: Gluecose content nmol/L = $\frac{\text{Determination OD --blacnk OD}}{\text{Standard OD --blank OD}}$ X Standard Concentration nmol/L X Sample dillution ratio

Fully automatic Gluecose content nmol/L = $\frac{\text{A determination}}{\text{A standard}}$ X Standard Concentration / X Sample dillution ratio

2). Tissue and cell calculation formulas (biochemical analyzers are not recommended for tissue and cells):

Microplate reader clorimetric: Gluecose content nmol/L = $\frac{\text{Determination OD --blacnk OD}}{\text{Standard OD --blank OD}}$ X Standard Concentration nmol/L ÷ The protein concentration of the sample

Note: The protein concentration determination kit is sold here (A045-2 Coomassie brilliant blue method, A045-3/-4 BCA method)

5.Product description: This kit can be used for the determination of glucose content in different samples, liquid samples (refer to the operation and calculation methods of serum), solid samples (refer to the operation and calculation methods of tissues and cells), mainly used for the determination of glucose content by microplate reader, human serum samples The reference range is (3.89-6.4mmol/L).

6. Performance indicators: 1. The absorbance of the reagent blank tube is less than or equal to 0.200 (optical diameter 1cm). 2. Linearity: in the range of 2.2 ~ 15mmol/L, r2>0.995. 3. Precision: CV≤3%, relative range between batches ≤5%. 4. Stability: The original kit should be stored at 2℃~ 8℃in the dark, and the validity period is 12 months. After opening, protect from light at 2℃~ 8℃Storage, stable for 1 month; reagents should not be frozen.

7, matters needing attention: 1. This product is only for scientific research, not for clinical diagnosis, and should not be taken. 2. If the content of the sample exceeds the upper limit of the detection range, the sample can be diluted with normal saline for determination, and the determination result is multiplied by the dilution factor. 3. The amount of reagents and samples can be increased or decreased in a ratio of 1:100 according to the requirements of the automatic biochemical analyzer.

References: 1. "Modern Clinical Biochemical Testing", Zhang Xiuming, Li Jianzhai, 2001, P84 2. "Practical Medical Laboratory Science", Zhu Zhongyong, 1992