

Glycogen Assay Kit

Note: Take two or three different samples for prediction before test.

Operation Equipment: Spectrophotometer/ microplate reader

Catalog Number: NA0598

Size:100T/96S

Components:

Extract reagent: 100mL×1, storage at 4°C.

Regent I: Powder×1, storage at 4°C.10 mg of glucose, add 1 mL of distilled water to dissolve it before use. Diluted with distilled water to 0.1 mg/mL glucose solution for standby, ready to use. 0.1mg/mL glucose standard solution, storage at 4°C.

Regent II: Powder×1, storage at 4°C.

Working solution: Pour 6 mL of distilled water into Reagent II and slowly pour 24 mL of concentrated sulfuric acid. Dissolve and mix thoroughly before use. Unused reagents are valid at 4°C for one week.

Product Description

Glycogen is a high molecular polysaccharide composed of glucose units. It is one of the main storage forms of sugar. It is mainly stored in the liver and muscle as backup energy, and is called liver glycogen and muscle glycogen, respectively. Glycogen can regulate blood glucose concentration. Glycogen can be synthesized in the liver when blood glucose rises. When blood sugar decreases, liver glycogen is broken down into glucose to supplement blood sugar. Therefore, liver glycogen is important to maintain the relative balance of blood sugar. Muscle glycogen is a form of glycogen storage in muscles. When lots of blood sugar is consumed during strenuous exercise, muscle glycogen cannot be broken down directly into blood sugar. It must first be broken down to produce lactic acid, which is circulated to the liver with the blood, and transformed into liver glycogen through glycogen glucose.

Determination principle: anthrone method. Glycogen is extracted with strong alkaline extract, and the glycogen content is measured using an anthrone method under strongly acidic conditions.

Reagents and Equipment Required but Not Provided.

Spectrophotometer/ microplate reader, desk centrifuge, transferpettor, micro glass cuvette/96-well plate, mortar, concentrated sulfuric acid (H₂SO₄) and distilled water.

Procedure:

I. Sample extraction:

1. Cells or bacteria: Collect 5-10 million bacteria or cells into a centrifuge tube, discard the supernatant after centrifugation; add 0.75mL of extraction reagent to ultrasonically break bacteria or cells (power 20% or 200W, ultrasonic 3s, 10s interval, repeat 30 times)); Transfer to a 10mL tube, boil in a boiling water bath for 20min (close tightly to prevent water loss), shake the tube every 5min to fully mix; take out the

tube and cool, take up to 5mL with distilled water and mix. Centrifuge at 8000g and 25°C for 10min, take the supernatant for testing.

2. Tissue: Weigh 0.1~0.2g sample and put it in a 10 mL tube, add 0.75 mL of Extraction solution, boil in boiling water bath for 20 min (close tightly to prevent water loss), shake the test tube every 5 min to fully mix. After all the tissue dissolved, take out the tube and cool down, then make up to 5 mL with distilled water. Centrifuge at 8000g and 25°C for 10 min, take the supernatant for testing.

II. Determination procedure:

1. Preheat the spectrophotometer/ microplate reader for 30 min, adjust wavelength to 620 nm, set zero with distilled water.

2. Sampling table (add the following reagents in EP tube)

Reagent(μL)	Blank Tube (A1)	Standard Tube (A2)	Test Tube (A3)
Sample			60
Reagent I		60	
distilled water	60		
Reagent II	240	240	240

Mix well, place in a boiling water bath for 10 minutes (close tightly to prevent water loss), cool, and take 200 μL into micro glass cuvette/96-well plate to read the absorbance of the blank tube, standard tube, and measurement tube at 620 nm, and record them as A1, A2, and A3. The blank tube and standard tube need only be tested once.

III. Calculation:

1. Sample weight

$$\begin{aligned} \text{Sorbitol (mg/g fresh weight)} &= (C_s \times V_1) \times (A_3 - A_1) \div (A_2 - A_1) \div (W \times V_1 \div V_2) \div 1.11 \\ &= 0.450 \times (A_3 - A_1) \div (A_2 - A_1) \div W \end{aligned}$$

2. Protein concentration

$$\begin{aligned} \text{Sorbitol (mg/mg prot)} &= (C_s \times V_1) \times (A_3 - A_1) \div (A_2 - A_1) \div (V_1 \times C_{pr}) \div 1.11 \\ &= 0.09 \times (A_3 - A_1) \div (A_2 - A_1) \div C_{pr} \end{aligned}$$

3. The number of bacteria or cells:

$$\begin{aligned} \text{Sorbitol (mg/10}^4 \text{ cell)} &= (C_s \times V_1) \times (A_3 - A_1) \div (A_2 - A_1) \div (\text{number of bacteria or cells} \times V_1 \div V_2) \div 1.11 \\ &= 0.450 \times (A_3 - A_1) \div (A_2 - A_1) \div \text{number of bacteria or cells} \end{aligned}$$

1.11: It is a constant that glucose content converted to glycogen content, That is, the color of 111 μg of glucose with anthrone reagent is equivalent to that of 100 μg of glycogen with anthrone reagent.

C_s: the concentration of standard, 0.1mg/mL

V₁: sample volume, 0.06 mL;

V₂: Total sample volume, 5 mL;

C_{pr}: sample protein concentration, mg/mL;

W: Sample weight, g

Number of bacteria or cells: 10⁴ as the unit, ten thousand

Note:

If A is greater than 1.4, dilute the sample with distilled water and multiply it by the corresponding dilution

factor in the calculation formula.

Recent Product Citations:

[1] Zheng J, Yu J, Jia M, et al. Indole enhances the survival of *Pantoea ananatis* YJ76 in face of starvation conditions[J]. *Journal of basic microbiology*, 2017, 57(7): 633-639.

[2] Xu L, Li Y, Yin L, et al. miR-125a-5p ameliorates hepatic glycolipid metabolism disorder in type 2 diabetes mellitus through targeting of STAT3[J]. *Theranostics*, 2018, 8(20): 5593.

[3] Ce Gu, Panpan Li, Wei Liu, et al. The role of insulin in transdifferentiated hepatocyte proliferation and function in serum-free medium. *Journal of Cellular and Molecular Medicine*. April 2019;(IF4.658)

References:

[1] Raunkjær K, Hvitved-Jacobsen T, Nielsen P H. Measurement of pools of protein, carbohydrate and lipid in domestic wastewater[J]. *Water research*, 1994, 28(2): 251-262.

[2] Carroll N V, Longley R W, Roe J H. The determination of glycogen in liver and muscle by use of anthrone reagent[J]. *J Biol Chem*, 1956, 220(2): 583-593.

Related Products:

NA0695/NA0454	Plant Tissue Fructose Content Assay Kit
NA0688/NA0447	Cellulase(CL) Activity Assay Kit
NA0841/NA0599	Trehalose Content Assay Kit
NA0691/NA0450	Trehalase Activity Assay Kit
NA0690/NA0449	Sorbitol Content Assay Kit
NA0689/NA0448	Sorbitol Dehydrogenase(SDH) Activity Assay Kit
NA0851/NA0609	Reducing Sugar(RS) Content Assay Kit
NA0693/NA0452	Blood Glucose Content Assay Kit

Technical Specification:

The detection limit: 0.002 mg/mL

The linear range: 0.003125-0.25 mg/mL