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Glucose Content Assay Kit

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

Operation Equipment: Spectrophotometer/microplate reader

Catalog Number: AK0219

Size:100T/96S

Components:

Solution I: 10 mL×1, 2 μmol/mL Glucose solution. Storage at 4℃.

Solution Ⅱ: Liquid 10 mL×1. Storage at 4℃.

Solution Ⅲ: Liquid 10 mL×1. Storage at 4℃.

Product Description

Glucose is not only the main substrate of cell energy metabolism, but also its metabolic intermediate is an important substrate of biosynthesis. Plants produce glucose through photosynthesis. In mammals, glucose is not only the sole source of energy for the nervous system, muscles and adipose tissue of the brain, but also is closely related to the synthesis of reductive coenzymes, lactose and milk fat.

Glucose oxidase catalyzes the oxidation of glucose to gluconic acid and hydrogen peroxide. Peroxidase catalyzes the oxidation of 4-aminoantipyrine bisphenol by hydrogen peroxide to form colored compounds with characteristic absorption peaks at 505 nm.

Reagents and Equipment Required but Not Provided.

Water-bath, transferpettor, spectrophotometer/microplate reader, micro glass cuvette/96 well flat-bottom plate, mortar/homogenizer and distilled water.

Procedure

a. Extraction of Glucose

Tissue treatment: The ratio of tissue mass (g) : distilled water volume (mL)=1:5 ~10. Suggest that weigh about 0.1 g of sample, add 1 mL distilled water and grind into homogenate. Boil them in a boiling water bath for 10 minutes (cover tightly to prevent water loss). After cooling, centrifuge them at room temperature for 10 min at 8000 g, then take the supernatant on standby.

b. Bacteria or cell treatment:

Collect the bacteria or cells into the centrifuge tube, discard the supernatant after centrifugation; According to the bacteria or cells (10^4) : distilled water volume (mL) is according the ratio of 500~1000: 1 (Recommend 1 mL of distilled water is added to 5 million bacteria or cells), ultrasonic broke bacteria or cells (ice bath, power of 20% or 200W, ultrasound for 3s, interval of 10s, repeat 30 times), set in a boiling water bath boil for 10 minutes (tightly closed to prevent moisture loss), after cooling, 8000 g, 25℃ centrifuge for 10 min, take supernatant on standby.

Measurement steps

a. Preheat the spectrophotometer/microplate reader for 30 min, adjust the wavelength to 505nm and adjust zero with distilled water.

b. Preparation of mixed reagent: mix Solution Ⅱ and Solution Ⅲ in equal volume (1:1) before use, prepare it fresh.

c. Add the following reagents successively into the 1.5ml centrifuge tube:

Reagent (μL)	Blank Tube	Standard Tube	Test Tube
Sample			20
Solution I		20	
ddH_2O	20		
mixed reagent	180	180	180

Mix thoroughly, incubate at 37℃ (mammals) or 25℃ (other species) in the water bath for 15 min, read the absorbance of wavelength at 505 nm . Note the light absorption values of blank tube, standard tube and measuring tube as A1, A2 and A3, respectively. Make one or two blank tube and one standard tube.

Calculation of glucose content:

1. Calculate by the protein concentration:

Glucose content (μmol/mg prot) $=(C_S \times V1)\times(A3-A1)\div(A2-A1)\div(V1\times Cpr)$

$=2\times(A3-A1)\div(A2-A1)\div Cpr$。

2. Calculate by Sample fresh weight:

Glucose content (μmol/g fresh weight) $= (C_S \times V1)\times(A3-A1)\div(A2-A1)\div(W\times V1\div V2)$

$=2\times(A3-A1)\div(A2-A1)\div W$

3. Calculate by the number of bacteria or cells

Glucose content (μmol/10^4 cell)$= (C_S \times V1)\times(A3-A1)\div(A2-A1)\div(500\times V1\div V2)$

$=0.004\times(A3-A1)\div(A2-A1)$

C_S: standard tube concentration, 2 μmol/mL;

V1: add sample volume, 20 μL=0.02 mL;

V2: total volume of the sample, 1 mL;

Cpr: sample protein concentration, mg/mL;

W: sample fresh weight, g;

500: total number of bacteria or cells, 5 million.

Note:

If the absorbance value of the sample is greater than 1.2, it is recommended to dilute the sample with distilled water for determination.

Recent Product Citations:

[1] Meixi Peng, Dan Yang, Yixuan Hou,et al. Intracellular citrate accumulation by oxidized ATM-mediated metabolism reprogramming via PFKP and CS enhances hypoxic breast cancer cell invasion. Cell Death and Disease. March 2019; (IF5.959)

[2] Jing Li,Yabing Duan,Chuanhong Bian,et al. Effects of validamycin in controlling Fusarium head blight caused by Fusarium graminearum: Inhibition of DON biosynthesis and induction of host resistance. Pesticide Biochemistry and Physiology. January 2019; 153:152- 160. (IF2.87)

References:

[1] Basagni U, Bonicolini F. Ready to use liquid reagent for determining the glucose content in blood: U.S. Patent 5,077,199[P]. 1991- 12-31.

[2] Kabasakalian P, Kalliney S, Westcott A. Enzymatic blood glucose determination by colorimetry of N, N-diethylaniline-4-aminoantipyrine[J]. Clinical chemistry, 1974, 20(5): 606-607.

Related Products:

AK0291/AK0290 Glucogen Content Assay Kit

AK0211/AK0210 Cellulase(CL) Activity Assay Kit

Technical Specifications:

The detection limit: 0.0188 μmol/mL

Linear range: 0. 125-8 μmol/mL