CREATIVE BIOMART®
Assay Kit

Glucose Assay Kit

Product Information

Common Name

Glucose

Cat.No.

Kit-0372

Product Overview

Glucose Assay Kit is a quantitative colorimetric glucose determination at 630 nm.

Description

Glucose (C6H12O6) is a ubiquitous fuel molecule in biology. It is oxidized through a series of enzyme-catalyzed reactions to form carbon dioxide and water, yielding the universal energy molecule ATP. Due to its importance in metabolism, glucose level is a key diagnostic parameter for many metabolic disorders. Increased glucose levels have been associated with diabetes mellitus, hyperactivity of thyroid, pituitary and adrenal glands. Decreased levels are found in insulin secreting tumors, myxedema, hypopituitarism and hypoadrenalism.

Applications

Direct Assays: glucose in biological samples (e.g. serum and plasma).
Drug Discovery/Pharmacology: effects of drugs on glucose metabolism.
Food and Beverages: glucose in food, beverages etc.

Usage

For research use only (RUO)

Storage

Store the reagent at room temperature and standard at -20°C, respectively. Shelf life: 12 months after receipt.

Kit Components

Reagent 50 mL
Standard 1 mL 300 mg/dL

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Materials Required but Not Supplied

Pipeting devices, centrifuge tubes, boiling water bath, tube holder.
Procedure using 96-well plate: Clear bottom 96-well plates (e.g. Corning Costar) and plate reader.
Procedure using cuvette: Spectrophotometer and Cuvets for measuring OD at 620-650nm

Detection method Colorimetric

Compatible Sample Types

Biological Sample, Plasma, Serum

Features & Benefits

Sensitive and accurate: Use as little as 5 µL samples. Linear detection range 0.7 mg/dL (39 µM) to 300 mg/dL (16.6 mM) glucose in 96-well plate.
Simple and convenient: The procedure involves addition of a single working reagent and incubation for 8 min in a boiling water bath.
Improved reagent stability: The optimized formulation has greatly enhanced the reagent and signal stability.
Low interference in biological samples. No pretreatments are needed. Assays can be directly performed on serum and plasma samples.

Assay Protocol

• Procedure using 96-well plate
1. Dilute standard in distilled water as follows.
No STD + H2O Vol (µL) Glucose (mg/dL)
1 150µL + 0µL 150 300
2 100µL + 50µL 150 200
3 50µL + 100µL 150 100
4 25µL + 125µL 150 50
5 0µL + 150µL 150 0
Set up 1.5-mL centrifuge tubes. Transfer 5 µL diluted standards and samples to appropriately labeled tubes. Transfer 500 µL Reagent to each tube. Close the tubes tightly and mix. Store diluted standards at -20°C for future use.

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2. Place the tubes in a tube holder and heat in a boiling water bath or heat block for 8 min. Cool down in cold water bath for 4 min.
3. Transfer 200 µL in duplicate into a clear bottom 96-well plate. Careful: avoid bubble formation. Read optical density at 620-650nm (peak absorbance at 630nm).
• Procedure using cuvette
1. Dilute standards and transfer 12 µL water blank, Standards and samples to appropriately labeled tubes.
Transfer 1200 µL Reagent to each tube. Close the tubes tightly and mix.
2. Place the tubes in a tube holder and heat in a boiling water bath for 8 min. Cool down in cold-water bath for 4 min.
3. Transfer 1000 µL reaction mixture into cuvet. Read optical density at 620-650nm (peak absorbance at 630nm) against blank.
Note:
If the Sample OD is higher than the Standard OD at 300mg/dL, dilute sample in water and repeat the assay.
To determine low glucose concentrations, use 50 µL sample and standards (instead of 5 µL) per 500 µL Reagent.

Analysis

Subtract blank OD (water, #5) from the standard OD values and plot the OD against standard concentrations.
Determine the slope using linear regression fitting. The glucose concentration of Sample is calculated as
= (ODSAMPLE – ODBLANK)/Slope(mg/dL)
ODSAMPLE and ODBLANK are optical density values of the sample and sample "Blank" (water or buffer in which the sample was diluted). Typical serum/plasma glucose values: 70 - 110 mg/dL.
Conversions: 1mg/dL glucose equals 55.5 µM, 0.001% or 10 ppm.