Glucose Assay Kit

Catalog No.: abx090673

Size: 100 tests (96 samples)

Detection Range: 0.05 mmol/L- 30 mmol/L

Sensitivity: 0.05 mmol/L

Storage: Store all components in the dark at 2 – 8 °C for up to 6 months.

Application: For detection and quantification of glucose concentration in serum, plasma, and other biological fluids.

Introduction

Glucose (or blood sugar) is the main source of energy in the body. An abnormal glucose level may lead to serious health complications. While a high blood sugar (or hyperglycemia) occurs when there is too much sugar in the blood because of insufficient insulin production or resistance to the actions of insulin in the body, a blood sugar level lower than the standard range (Hypoglycemia) is often related to diabetes treatment. The high blood sugar is a symptom that characterizes diabetes and increases the risk for heart diseases, kidney diseases and nerve damage.

Abbexa's Glucose Assay Kit is a quick, convenient, and sensitive method for measuring and calculating Glucose content and is based on a Glucose Oxidase-Peroxidase Method. Through the action of peroxidase, glucose is converted to glucanic acid and H2O2 by glucose oxidase. H2O2 reacts with 4-amino phenazone and phenol to form a quinone, which can be measured by a microplate reader or spectrophotometer at a wavelength of 505 nm.

Kit components

1. Phenol Solution: 2 × 60 mL
2. Enzyme Solution: 2 × 60 mL
3. Glucose Standard (5 mmol/L): 1 vial

Materials Required But Not Provided

1. Microplate reader or spectrophotometer (505 nm)
2. Double distilled water
3. Normal saline (0.9% NaCl)
4. Pipette and pipette tips
5. Incubator
6. Vials/tubes
7. Vortex mixer

Protocol

A. Preparation of samples and reagents

The following sample preparation methods are intended as a guide and may be adjusted as required depending on the specific samples used.

- **Serum:** Samples should be collected into a serum separator tube. Coagulate the serum by leaving the tube undisturbed in a vertical position overnight at 4°C or at room temperature for up to 1 hr. Centrifuge at approximately 2000 × g for 15 mins at 4°C. If a precipitate appears, centrifuge again. Take the supernatant, keep on ice and assay immediately, or aliquot and store at -80°C for up to 1 month.
- **Plasma:** Collect plasma using heparin as the anticoagulant. Centrifuge for 10 mins at 1000-2000 x g at 4°C, within 30 mins of collection. If precipitate appears, centrifuge again. Avoid hemolytic samples. Take the supernatant (avoid taking the middle layer containing white blood cells and platelets), keep on ice and assay immediately, or aliquot and store at -80°C for up to 1 month.

Samples should not contain detergents such as SDS, Tween-20, NP-40 and Triton X-100, or reducing agents such as DTT.

It is recommended to carry out a preliminary experiment to determine the optimal dilution factor of samples before carrying out the formal experiment. Dilute samples into different concentrations with normal saline (0.9% NaCl) or PBS (0.01 M, pH 7.4), then carry out the assay procedure.

Note:

- Fresh samples or recently obtained samples are recommended to prevent degradation and denaturalization that may lead to erroneous results.
- The recommended dilution factor for Rat and Human Plasma, and Mouse and Human Serum sample is 1.

2. Reagents

- **Enzyme working solution:** Mix Phenol solution and enzyme solution at 1:1 ratio. Prepare immediately before carrying out the assay.

B. Assay Procedure

1. Set the Blank, Standard, and Sample tubes.
2. Add 2000 µl of freshly prepared enzyme working solution in each tube.
3. Add 20 µl of double distilled water in Blank tube.
4. Add 20 µl of Glucose Standard in the Standard tube.
5. Add 20 µl of Sample to the sample tube.
6. For each tube, mix fully and incubate at 37°C for 25 minutes.
7. Measure the OD values at 505 nm with a microplate reader.

C. Calculation of Results

$$\text{Glucose (mmol/L)} = \frac{\Delta A_1}{\Delta A_2} \times c \times f$$

where:

ΔA_1	OD value of the sample ($OD_{Sample} - OD_{Blank}$)
ΔA_2	OD value of the Standard ($OD_{Standard} - OD_{Blank}$)
c	concentration of standard (5 mmol/L)
f	dilution factor of the sample before carrying out the assay