

# **Glucose Assay Kit**

Glucose Assay Kit is a detection kit for the quantification of Glucose Content in serum, plasma, urine, saliva, milk, culture medium, food and beverages sample.

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Package: 100 tests

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#### INTRODUCTION

Glucose is a sugar with the molecular formula C6H12O6, which is often abbreviated as Glc. It is overall the most abundant monosaccharide, a subcategory of carbohydrates. It is mainly made by plants and most algae during photosynthesis from water and carbon dioxide, using energy from sunlight. It is used by plants to make cellulose, the most abundant carbohydrate in the world, for use in cell walls, and by all living organisms to make adenosine triphosphate (ATP), which is used by the cell as energy.

#### PRINCIPLE OF THE ASSAY

This Glucose Assay Kit is a detection kit for the quantification of Glucose Content in serum, plasma, urine, saliva, milk, culture medium, food and beverages sample. This assay uses a single Working Reagent that combines the reaction and color reaction in one step. The change in color intensity of the reaction product at O.D. 565 nm is directly proportional to Glucose in the sample.

## **MATERIALS PROVIDED & STORAGE INFORMATION**

The kit is shipped on ice. Store all components at-20°C upon receiving. Shelf life: 6 months after receipt.

Component	Quantity	Storage information
Assay Buffer	10 mL	-20°C
NAD/MTT	1 mL	-20°C
GDH	120 μL	-20°C
Diaphorase	120 μL	-20°C
Glucose Standard (300 mg/dL)	1 mL	-20°C

# MATERIALS REQUIRED BUT NOT PROVIDED

- Microplate reader capable of reading at O.D. 565 nm
- Clear flat-bottom 96 well microplate
- Centrifuge and centrifuge tube
- Deionized or Distilled water
- Pipettes, pipette tips and Multichannel micropipette reservoir

## **TECHNICAL NOTES AND PRECAUTIONS**

- Wear protective gloves, clothing, eye, and face protection especially while handling blood or body fluid samples.
- Reagents are for research use only. Normal precautions for laboratory reagents should be exercised while using the reagents. Please refer to Material Safety Data Sheet for detailed information.
- All reagents should be mixed by gentle inversion or swirling prior to use.
  Do not induce foaming.
- Before using the kit, spin tubes and bring down all components to the bottom of tubes.
- It is highly recommended assaying the Standards and samples in duplicates.
- Change pipette tips between the addition of different reagent or samples.

# SAMPLE COLLECTION & STORAGE INFORMATION

The sample collection and storage conditions listed below are intended as general guidelines. Sample stability has not been evaluated.

<u>Milk</u>: Mixing 600  $\mu$ L milk with 100  $\mu$ L 6 M HCl and centrifuge sample (5 minutes at 14,000 rpm) and transfer supernatant into a clean tube. Add 170  $\mu$ L 6 M NaOH for 1 mL supernatant, and centrifuge sample (5 minutes at 14,000 rpm). Using the clear supernatant for the assay.

<u>Saliva</u>: Centrifuge sample (5 minutes at 14,000 rpm) and use the clear supernatant for the assay.

#### Note:

All samples can be stored at-20°C. Avoid repeated freeze-thaw cycles.

#### REAGENT PREPARATION

- Working Reagent: For each well, mixing 80 μL Assay Buffer, 1 μL GDH, 1 μL
  Diaphorase and 8 μL NAD/MTT. Fresh reconstitution is recommended.
- Standards: Diluted 48  $\mu$ L standard (300 mg/dL, 16.7 mM) with 352  $\mu$ L distilled water to yield 2 mM. Dilute standard in dH2O as follows:

Standard	Glucose (mM)	2 mM STD (μL)	ddH20 (μL)
1	2.0	200	0
2	1.2	120	80
3	0.6	60	140
4	0	0	200

Note: Working Reagent is stable for 2 hours.

# **ASSAY PROCEDURE**

Equilibrate all components to room temperature. During experiment, keep thawed Enzymes in a refrigerator or on ice. Briefly centrifuge tubes before use.

	Standard well	Sample well	Blank well
Standards	20 μL	-	-
Each Sample	-	20 μL	-
Distilled water	-	-	20 μL
Working Reagent	80 μL	80 μL	80 μL

Tap plate to mix immediately. Incubate for **30 minutes** at **room temperature**.

Read the absorbance at O.D. 565 nm.

# **CALCULATION OF RESULTS**

1. Glucose concentration of a sample is calculated as:

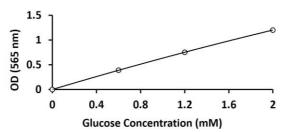
Glucose (mM) = 
$$[(OD_{Sample} - OD_{Blank}) / Slope] \times n$$

Note:

- OD<sub>Sample</sub> and OD<sub>Blank</sub>: the O.D. 565 nm values of the Sample and blank.
- > n: the sample dilution factor.
- 2. If the Glucose Sample concentration is higher than 2 mM, dilute sample in water and repeat the assay. Multiply result by the dilution factor.

#### **EXAMPLE OF TYPICAL STANDARD CURVE**

The following figures demonstrate typical results with the Glucose Assay Kit. One should use the data below for reference only. This data should not be used to interpret actual results.



# **QUALITY ASSURANCE**

# Sensitivity

0.03 mM