

Description

A colorimetric assay kit for the quantitative determination of glucose in serum, plasma, urine, saliva, milk, culture medium, food and beverages. The kit uses an enzyme to reduce NAD; the produced NADH, measured at 340 nm, is proportional to the glucose concentration, with a linear detection range of 0.1 to 3 mM in a 96-well plate.

Specifications

Detection Method	Colorimetric (OD340 nm)
Detection Range	0.1 to 3 mM (1.8 to 54 mg/dL) glucose (96-well plate)
Sample Type	Serum,Plasma,Urine,Saliva,Milk,Culture medium,Food,Beverages
Species Reactivity	All
Assay Time	20 min
Kit Size	100 Assays
Equipment Required	Microplate reader
Storage	-20°C
Shelf Life	6 months
Shipping	Gel Pack

Kit Components

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

Ordering Information

Assay Genie Cat. No.	Pack Size
BA0242	100 Assays

Assay Procedure

Note: This is a sample protocol. Protocols are batch/lot specific - always follow the instructions supplied with your kit.

1. Equilibrate all components to room temperature. During the experiment, keep thawed GDH in a refrigerator or on ice.
2. Standards and samples: for the 3 mM standard, mix 72 µL 300 mg/dL standard with 328 µL dH₂O, then dilute in dH₂O to give 3.0, 1.8, 0.9 and 0 mM points (see Standard Dilution table). Transfer 20 µL standards and samples into separate wells.
3. Prepare sufficient Working Reagent by mixing, per well, 80 µL Assay Buffer, 1 µL GDH and 8 µL NAD in a clean tube. Transfer 80 µL Working Reagent into each reaction well and tap the plate briefly and thoroughly to mix.
4. Incubate 20 min at room temperature. Read optical density at 340 nm.

Standard Dilution (Glucose)

No	3 mM STD + H ₂ O	Vol (µL)	Glucose (mM)
1	200 µL + 0 µL	200	3.0

No	3 mM STD + H2O	Vol (µL)	Glucose (mM)
2	120 µL + 80 µL	200	1.8
3	60 µL + 140 µL	200	0.9
4	0 µL + 200 µL	200	0

Calculation

Subtract the blank OD (water, #4) from the standard OD values and plot ΔOD against standard concentrations. Determine the slope and calculate: $[Glucose] = (OD_SAMPLE - OD_BLANK) / Slope \times n$ (mM), where OD_SAMPLE and OD_BLANK are the optical density values of the sample and water respectively and n is the sample dilution factor. Conversions: 1 mg/dL glucose equals 55.5 µM, 0.001% or 10 ppm. If the calculated glucose concentration exceeds 3 mM, dilute the sample in water and repeat, multiplying the result by the dilution factor.

Notes

- Saliva samples should be centrifuged for 5 min at 14,000 rpm prior to assay. Milk samples should be cleared by mixing 100 µL 6 M HCl with 600 µL milk, centrifuging 5 min at 14,000 rpm, transferring the supernatant, adding 170 µL 6 M NaOH per mL supernatant, mixing and centrifuging again (dilution factor n = 1.36).
- Samples can be analysed immediately after collection or stored in aliquots at -20°C. Avoid repeated freeze-thaw cycles. If particulates are present, centrifuge and use the clear supernatant.
- To determine glucose in phenol-red culture medium, dilute both sample and glucose standards in the same glucose-free medium for the colorimetric assay.
- Reagents are for research use only. Refer to the MSDS for detailed safety information.

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Technical specifications are based on manufacturer documentation and may be updated. Protocols shown are summaries - always follow the instructions supplied with your kit/lot.