PATHOZYME DIAGNOSTICS

LIQVIPATH GLUCOSE-*HK*

(Hexokinase Method)

CAT NO.: GHK

Reagent kit for quantitative estimation of Glucose in Serum or Plasma or Urine or CSF.

DIAGNOSTIC SIGNIFICANCE:

Serum/Plasma Glucose levels may be abnormally high (hyperglycemia) or abnormally low (hypoglycemia). 1 Glucose measurements are used in the diagnosis and treatment of carbohydrate metabolism disorders including diabetes mellitus, neonatal hypoglycemia, and idiopathic hypoglycemia, and of pancreatic islet cell carcinoma.

Glucosuria (the presence of urinary glucose) is common in healthy, pregnant women. The cardinal feature of the glucosuria of pregnancy is a conspicuous variability both from day to day and during the course of the day. 2 Glucose is not present in normal patient urine.

Determinations of cerebrospinal fluid (CSF) glucose helps distinguish bacterial from viral meningitis; the glucose value is often low (less than 40% to 45% of simultaneously analyzed, equilibrated serum glucose) in bacterial meningitis and tuberculous meningitis and is generally normal in viral disease. Carcinomatous meningitis (widespread infiltration of the meninges by tumor cells) also drives CSF glucose values below the normal range.

PRINCIPLE:

Glucose is phosphorylated by Hexokinase (HK) in the presence of adenosine triphosphate (ATP) and magnesium ions to produce Glucose-6-phosphate (G-6-P) and adenosine diphosphate (ADP). Glucose-6-phosphate dehydrogenase (G6P-DH) specifically Oxidizes G-6-P to 6-phosphogluconate with the concurrent reduction of nicotinamide adenine dinucleotide phosphate (NADP) to nicotinamide adenine dinucleotide phposphate reduced (NADPH). The change in absorbance at 340nm is proportional to the amount of Glucose present in the sample.

$$\text{ATP + Glucose} \xrightarrow{\text{HK}} \text{G-6-P + ADP}$$

$$\text{G-6-P + NADP} \xrightarrow{\text{G6PDH}} \text{D-Gluconate-6-phosphate + NADPH + H}^{+}$$

SPECIMEN COLLECTION:

Fasting serum or plasma (heparin or sodium fluoride) samples, free from hemolysis, are the recommended specimens. Separate from red cells rapidly to minimize loss of glucose through glycolysis. Fresh, random collections are recommended for urine specimens.

KIT PRESENTATION:

PACK SIZE	2 X 50 ml	4 X 50 ml	5 X 100 ml
R1-Glucose-HK	2 X 40 ml	4 X 40 ml	4 X 100 ml
R2-Glucose-HK	2 X 10 ml	2 X 20 ml	2 X 52 ml
Glucose Standard	1 X 2 ml	1 X 2 ml	1 X 5 ml

WORKING REAGENT PREPARATION:

Mixing 4 volumes of **R1-GlucoseHK** with 1 volume of **R2 GlucoseHK.**

The working reagent is stable for **30** days at 2-8°C.

REAGENT STORAGE AND STABILITY:

GlucoseHK Reagents and Standard are stable at 2-8°C until the expiry date stated on the label.

ASSAY PARAMETERS:

Reaction	**: End Point**	**Sample Volume**	**: 10 µl**
Wavelength	**: 340 nm**	**R1 + R2 Volume**	**: 800 µl + 200 µl**
Flow Cell Temp.	**: 37°C**	**Std. Conc.**	**: 100 mg/dl**
Incubation Time	**: 5 Minutes**	**Zero Setting**	**: Reagent**
Incubation Temp.	**: 37°C**	**Linearity**	**: 500 mg/dl**

PROCEDURE:

Pipette into TT	Blank	Standard	Test
R1-Glucose-HK	800 µl	800 µl	800 µl
Glucose Std (100mg/dl)	--	10 µl	--
Sample (Test)	--	--	10 µl
R2-Glucose-HK	200 µl	200 µl	200 µl

Mix & incubate at 37°C for 5 minutes. Read absorbance of standard (**S**) and Test (**T**) after 5 minutes against reagent blank at 340 nm.

CALCULATION:

Glucose (mg/dl) = Abs T ÷ Abs S X 100

Conversion factor

Glucose (mg/dl) X 0.05551= Glucose (mmol/L)

NORMAL VALUES:

Serum / Plasma : 74 - 106 mg/dl (4.10 - 5.90 mmol/L)
Cerebral Spinal fluid : 40 - 70 mg/dl (2.20 - 3.90 mmol/L)
Urine : ≤ 15 mg/dl (0.84 mmol/L) *

*** (Value is based on an average quantity of urine of 1350 ml/day)**

Expected values may vary with age, sex, diet and geographical location. Each laboratory should determine its own expected values as dictated by good laboratory practice.

LINEARITY:

This method is linear up to **500 mg/dl**. For values above 500 mg/dl, dilute the sample suitably with 0.9 % saline, and repeat the assay. Apply dilution factor to obtain final result.

REFERENCES:

1. Burrin JM, Price CP. Measurement of blood glucose. Ann. Clin. Biochem. **22**, (1985), 327
2. Tietz. Fundamentals of Clinical Chemistry, Chap. 23.447 (2001).
3. Young, D.S., Effects of Drugs on Clinical Laboratory Tests, AACC Press, 5th Edition 2000.
4. CLSI/NCCLS Evaluation Protocol EP5-A, 1999.

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Expiry Date | IVD In-Vitro Diagnostics Use | Storage | Mfg. Date | LOT Batch Number | REF Catalogue Number | See Package Insert

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