Glucose Hexokinase FS*

Diagnostic reagent for quantitative in vitro determination of glucose in serum, plasma or urine on photometric systems

Order Information

Cat. No.	Kit size						
1 2511 99 10 021	R1	5 x	20 mL	+	R2	1 x	25 mL
1 2511 99 10 026	R1	5 x	80 mL	+	R2	1 x	100 mL
1 2511 99 10 023	R1	1 x	800 mL	+	R2	1 x	200 mL
1 2511 99 10 704	R1	8 x	50 mL	+	R2	8 x	12.5 mL
1 2511 99 10 917	R1	8 x	60 mL	+	R2	8 x	15 mL

Summary [1,2]

Measurement of glucose concentration in serum or plasma is mainly used in diagnosis and monitoring of treatment in diabetes mellitus. Other applications are the detection of neonatal hypoglycemia, the exclusion of pancreatic islet cell carcinoma as well as the evaluation of carbohydrate metabolism in various diseases.

Method

Enzymatic UV test using hexokinase

Principle

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

$$\text{Glucose-6-phosphate} + \text{NAD}^+ \xrightarrow{\text{G6P-DH}} \text{Gluconate-6-P} + \text{NADH} + \text{H}^+$$

Reagents

Components and Concentrations

R1:	TRIS buffer	pH 7.8	100 mmol/L
	Mg^{2+}		4 mmol/L
	ATP		2.1 mmol/L
	NAD		2.1 mmol/L
R2:	Mg^{2+}		4 mmol/L
	Hexokinase (HK)		≥ 7.5 kU/L
	Glucose-6-phosphatedehydrogenase (G6P-DH)		≥ 7.5 kU/L

Storage Instructions and Reagent Stability

Reagents are stable up to the end of the indicated month of expiry, if stored at 2 – 8°C, protected from light and contamination is avoided. Do not freeze the reagents!

Warnings and Precautions

1. The reagents contain sodium azide (0.95 g/L) as preservative. Do not swallow! Avoid contact with skin and mucous membranes.
2. Reagent 2 contains animal material. Handle the product as potentially infectious according to universal precautions and good laboratory practice.
3. In very rare cases, samples of patients with gammopathy might give false results [6].
4. Please refer to the safety data sheets and take the necessary precautions for the use of laboratory reagents. For diagnostic purposes, the results should always be assessed with the patient's medical history, clinical examinations and other findings.
5. For professional use only!

Waste Management

Please refer to local legal requirements.

Materials required but not provided

NaCl solution 9 g/L
General laboratory equipment

Reagent Preparation

Reagents are ready to use.

Specimen

Serum, plasma or urine
Separate from cellular content at the latest 1h after blood collection.
Stability in plasma after addition of a glycolytic inhibitor (fluoride, monoiodacetate, mannose) [3]:

2 days	at	20 – 25°C
7 days	at	4 – 8°C
1 day	at	–20°C

Stability in serum (separated from cellular contents, hemolysis free) without adding a glycolytic inhibitor [2,4]:

8 h	at	25°C
72 h	at	4°C

Stability in urine [3]:

2 h	at	20 – 25°C
2 h	at	4 – 8°C

Only freeze once!

Discard contaminated specimens!

Assay Procedure

Application sheets for automated systems are available on request.

Wavelength	340 nm, Hg 334 nm, Hg 365 nm
Optical path	1 cm
Temperature	20 – 25 °C/37 °C
Measurement	Against reagent blank

	Blank	**Sample/Calibrator**
Sample/Calibrator	-	10 µL
Dist. water	10 µL	-
Reagent 1	1000 µL	1000 µL
Mix and incubate 1–5 min. at 20 – 25 °C/37 °C. Read absorbance A1, then add:		
Reagent 2	250 µL	250 µL
Mix, incubate 5 min. at 37 °C or 10 min. at 20 – 25 °C. Read absorbance A2 against reagent blank within 30 min.		

$\Delta A = (A2 - A1)$ Sample/Calibrator

Calculation

With factor

Multiply ΔA by the corresponding factor f from the table below in order to calculate the glucose concentration.

Wavelength	f [mg/dL]	f [mmol/L]
340 nm	361	20.0
Hg 334 nm	367	20.5
Hg 365 nm	667	37.1

With calibrator

$$\text{Glucose [mg/dL]} = \frac{\Delta A \text{ Sample}}{\Delta A \text{ Cal.}} \times \text{Conc. Cal. [mg/dL]}$$

Conversion factor

Glucose [mg/dL] x 0.05551= Glucose [mmol/L]

* fluid stable

Calibrators and Controls

For calibration of automated photometric systems, DiaSys TruCal U calibrator is recommended. The calibration values of this calibrator have been made traceable to the reference method gas chromatography – isotope dilution mass spectrometry (GC-IDMS). Glucose Standard FS may be used alternatively for calibration. For internal quality control DiaSys TruLab N, P and TruLab Urine controls should be assayed. Each laboratory should establish corrective action in case of deviations in control recovery.

	Cat. No.	Kit size
TruCal U	5 9100 99 10 063	20 x 3 mL
	5 9100 99 10 064	6 x 3 mL
TruLab N	5 9000 99 10 062	20 x 5 mL
	5 9000 99 10 061	6 x 5 mL
TruLab P	5 9050 99 10 062	20 x 5 mL
	5 9050 99 10 061	6 x 5 mL
TruLab Urine Level 1	5 9170 99 10 062	20 x 5 mL
	5 9170 99 10 061	6 x 5 mL
TruLab Urine Level 2	5 9180 99 10 062	20 x 5 mL
	5 9180 99 10 061	6 x 5 mL
Glucose Standard FS	1 2500 99 10 030	6 x 3 mL

Performance Characteristics

Measuring range

The test has been developed to determine glucose concentrations within a measuring range from 2 – 900 mg/dL (0.1 – 50 mmol/L) measured at 365 nm, respectively within a measuring range from 2 – 500 mg/dL (0.1 – 28 mmol/L) measured at 334/340 nm.
When values exceed these ranges serum and plasma samples should be diluted 1+2 with NaCl solution (9 g/L) and the result multiplied by 3, urine samples should be diluted 1+10 with dist. water and the results multiplied by 11.

Specificity/Interferences

No interference was observed by ascorbic acid up to 30 mg/dL, bilirubin up to 40 mg/dL, hemoglobin up to 500 mg/dL and lipemia up to 2000 mg/dL triglycerides, when worked with substrate start. For further information on interfering substances refer to Young DS [5].

Sensitivity/Limit of Detection

The lower limit of detection is 2 mg/dL (0.1 mmol/L).

Precision (at 37 °C)

Intra-assay precision n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	65.7	1.39	2.11
Sample 2	121	2.54	2.11
Sample 3	298	6.57	2.21

Inter-assay precision n = 20	Mean [mg/dL]	SD [mg/dL]	CV [%]
Sample 1	91.0	0.86	0.94
Sample 2	117	1.07	0.91
Sample 3	290	2.28	0.79

Method Comparison

A comparison of DiaSys Glucose Hexokinase FS (y) with a commercially available test (x) using 73 samples gave following results:
y = 1.00 x + 0.00 mg/dL; r = 0.998

Reference Range [1]

	[mg/dL]	[mmol/L]
Newborns:		
Cord blood	63 – 158	3.5 – 8.8
1 h	36 – 99	2.0 – 5.5
2 h	36 – 89	2.2 – 4.9
5 – 14 h	34 – 77	1.9 – 4.3
10 – 28 h	46 – 81	2.6 – 4.5
44 – 52 h	48 – 79	2.7 – 4.4
Children (fasting):		
1 – 6 years	74 – 127	4.1 – 7.0
7 – 19 years	70 – 106	3.9 – 5.9
Adults (fasting):		
Serum/plasma	70 – 115	3.9 – 6.4

Urine: ≤ 15 mg/dL (0.84 mmol/L)

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

Each laboratory should check if the reference ranges are transferable to its own patient population and determine own reference ranges if necessary.

Literature

1. Thomas L. Clinical Laboratory Diagnostics. 1st ed. Frankfurt: TH-Books Verlagsgesellschaft; 1998. p. 131-7, 1368.
2. Sacks DB. Carbohydrates. In: Burtis CA, Ashwood ER, editors. Tietz Textbook of Clinical Chemistry. 3rd ed. Philadelphia: W.B Saunders Company; 1999. p. 750-808.
3. Guder WG, Zawta B et al. The Quality of Diagnostic Samples. 1st ed. Darmstadt: GIT Verlag; 2001; p. 30-1, 50-1.
4. Sacks DB, Bruns DE, Goldstein DE, Mac Laren NK, Mc Donald JM, Parrott M. Guidelines and recommendations for laboratory analysis in the diagnosis and management of diabetes mellitus. Clin Chem 2002; 48: 436-72.
5. Young DS. Effects of Drugs on Clinical Laboratory Tests. 5th ed. Volume 1 and 2. Washington, DC: The American Association for Clinical Chemistry Press 2000.
6. Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. ClinChemLabMed 2007;45(9):1240-1243.

Manufacturer

IVD CE

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