المصنع المتحد للكواشف الطبية
ص.ب ٩٤٦٦ - الدمام ٣١٤١٣ - المملكة العربية السعودية
تلفون : ٢٠٠٤ ٨١٢ - ٨١٢ ١٢٣٣ (٠٣) - فاكس : ١٧٠٤ ٨١٢ (٠٣) - ترخيص رقم ١٩٨٠ / ص

United Diagnostics Industry
P.O. Box 9466 - Dammam 31413 - K.S.A.
Tel : (03) 812 1233 - 812 2004 - Fax: (03) 812 1704
Lic No. 1980 /S www.udignost.com

GLUCOSE (OX.) LIQUID COLORIMETRIC

MODIFIED TRINDER / GOD METHOD

REF 037L

FOR IN VITRO DIAGNOSTIC USE

INTENDED USE

Quantitative determination of glucose in serum or plasma using a modified glucose oxidase / Trinder method.

DIAGNOSTIC SIGNIFICANCE

Glucose is a major carbohydrate present in the peripheral blood. The oxidation of glucose is the major source of cellular energy in the body. Glucose determinations are run primarily to aid in the diagnosis and treatment of diabetes-mellitus. Elevated glucose levels may be associated with pancreatitis, pituitary or thyroid dysfunction, renal failure and liver disease, whereas low glucose levels may be associated with insulinoma, hypopituitarism, neospasms, or insulin hypoglycemia[1, 2].

RANGE OF EXPECTED VALUES IN SERUM[3]

70-105 mg/dL (3.9-5.8 mmol/L) (FASTING)

METHOD PRINCIPLE

The enzymatic reaction sequence employed in the assay of glucose is as follows:

$$\text{Glucose} + O_2 + H_2O \xrightarrow{\text{GOD}} \text{Gluconic Acid} + H_2O_2$$

$$2H_2O_2 + 4\ \text{Aminoantipyrine} + \text{PHBS} \xrightarrow{\text{POD}} \underset{\text{(Red Color)}}{\text{Quinoneimine dye}} + 4H_2O$$

D-glucose, in the presence of glucose oxidase and oxygen, is oxidized to gluconic acid and hydrogen peroxide. The hydrogen peroxide is oxidatively coupled with 4-aminoantipyrine and p-hydroxybenzene sulfonate in the presence of peroxidase to form a stable red quinoneimine dye. The quinoneimine dye has an absorption maximum at 510 nm. The amount of color produced is directly proportional to the glucose content of the sample up to a concentration of 500 mg/dL The UDI procedure is modified Trinder[4, 5] method.

REAGENTS

1. ***GLUCOSE (OX.) LIQUID REAGENT*** : 15 U/ml glucose oxidase, 1.2 U/ml peroxidase, 0.38 Mm 4-aminoantipyrine, 10 mM p-hydroxybenzene sulfonate, non-reactive stabilizers and fillers in buffer. Keep tightly capped and protected from contamination. The reagent can be used until the expiration date indicated on the bottle label.
2. ***GLUCOSE STANDARD (100 mg/dL or 5.55 mmol/L)*** : A solution containing 0.100% w/v D-glucose and preservative. Must be kept tightly capped and protected from contamination. Can be used until the expiration date indicated on the bottle label.

REAGENT STORAGE & STABILITY

1. Glucose (Ox.) Liquid Reagent and standard are stable up to expiration date indicated on the bottle label when stored at 2-8 °C. Once opened, use within one month.

CHEMICAL PRECAUTIONS

Exercise the normal precautions required for the handling of all laboratory reagents. Pipetting by mouth is not recommended for any laboratory reagent.

INDICATIONS OF REAGENT DETERIORATION

1. Physical appearance
   Turbidity has occurred. Turbidity may be a sign of contamination.
2. Control assays
   Failure to obtain accurate results in the assay of control materials may indicate reagent deterioration.

NOTE: UDI cannot guarantee the stability of reagents which have been:

a) transferred from their original containers
b) improperly stored
c) contaminated during use.

SPECIMEN COLLECTION AND HANDLING

1. Serum
   Collect whole blood by venipuncture and allow to clot (normally takes about 20 minutes). Centrifuge and remove serum. If the serum is permitted to remain in contact with the clot, glycolysis will occur. The glucose loss due to glycolysis is about 10-20 mg/dL/hr at 37 °C and about 5-10 mg/dL/hr at room temperature[6]. The rate of glycolysis is greater in newborn infants than in adults. Non-hemolyzed, cell free serum is usually stable for 24 hours at 2-8 °C.
2. Plasma [6]
   Collect specimen by venipuncture and separate plasma from cells within 20 minutes to avoid the effects of glycolysis. The following concentration of common anti-coagulants may be used. Oxalate (<50 mg/ml blood), fluoride (<10 mg/ml blood), EDTA (<10mg/ml blood), Citrate (<50 mg/ml blood), heparin (<2 mg/ml blood), thymol (<1 mg/ml blood), sodium or potassium monoiodoacetate (<2 mg/ml blood).

MATERIALS PROVIDED

Glucose (Ox.) Liquid Reagent and Glucose Standard (100 mg/dL or 5.55 mmol/L)

ADDITIONAL MATERIALS REQUIRED, BUT NOT PROVIDED

Reagent and sample pipettes, Test Vials or Cuvettes, Timer, Test tube rack, 37 °C heating bath, Control serum, Spectrophotometer.

PROCEDURE (AUTOMATED)

Refer to specific instrument application mannual available from us.

PROCEDURE (MANUAL)

Pipette into clean cuvettes

	REAGENT BLANK	STANDARD	TEST
Glucose (Ox.) Liquid Rgnt	1.0 ml	1.0 ml	1.0 ml
Pre-warm at 37 °C and add:			
Glucose Standard	- -	0.01 ml	- -
Sample	- -	- -	0.01 ml
Mix and incubate at 37 °C for 10 min. Read the absorbance of sample and standard at 510 ± 10 nm against Reagent Blank.			

STABILITY OF ENDPOINT REACTION

The final color developed in the reaction is stable for at least 30 minutes when kept at 37°C or lower; However, direct sunlight must be avoided.

CALCULATION OF RESULTS

Use the absorbance measurements of the STANDARD and TEST(S) to calculate Glucose values as follows: (A = Absorbance)

$$\frac{A(TEST)}{A(STANDARD)} \times \text{Conc. of STANDARD (mg/dL)} = \text{Glucose Conc.in TEST (mg/dL)}$$

Example : Assume the value of the STANDARD to be 100 mg/dl (5.55 mmol/L), and that it gave an absorbance of 0.325 while the TEST gave an abosrbance of 0.300. The glucose concentration of the TEST may then be calculated as follows::

$$\frac{0.300}{0.325} \times 100 \text{ mg/dl} = 92 \text{ mg/dl} \quad \text{OR } 5.12 \text{ mmol/L}$$

PROCEDURE LIMITATIONS

Hemolyzed samples should not be used.

The following substances reportedly cause decreased glucose values

Ascorbic Acid when greater than 5 mg/dL, Hemoglobin when greater than 0.7 g/dL, Bilirubin when greater than 15 mg/dL, Uric Acid when greater than 2.0 mg/dL, Cysteine when greater than 10 mg/dL, Acetylsalicylic Acid, L-DOPA, Tetracycline, Mercurial diuretics, and Gentisic Acid.

For a more comprehensive listing of drug effects on laboratory tests refer to reference 7.

PERFORMANCE CHARACTERISTICS

1. LINEARITY & SENSITIVITY

Glucose values up to 500 mg/dL can be measured with this method. Typically, 0.001 a represents about 0.3 mg glucose/dL when measured in a 1-cm light path.

2. COMPARISON : UDI reagents tested on MANUAL SYSTEMS(y) was compared with similar UDI reagent for other systems (x) which in turn is matching with CAPS survey results . The systematic difference between the results were within CLIA specified limits, N =25

Correlation Coefficient 0.99

Regression Equation $y = x + 6.98$

3. PRECISION:

	Mean mg/dL	SD	CV%
Within run	93.8	1.94	2.07
Run to run	283	8.64	3.05.

PROCEDURE NOTES

A sample blank must be determined with moderately icteric and lipemic sera as follows:

1. Add 20 µl (0.02 ml) of sample to 2.0 ml of 0.85% sodium chloride (saline) and mix thoroughly.
2. Zero the photometer at 510 nm with 0.85% sodium chloride. Blank
3. Read and record absorbance of sample.
4. Subtract this absorbance value from that obtained from MANUAL ENDPOINT PROCEDURE and calculate according to above instructions.

QUALITY CONTROL

For accuracy and precision check, we recommend use of normal and abnormal UDI controls based on human serum.

ORDERING INFORMATION:

UDITROL 'N' (Normal Serum Control) REF # 070N-010 2x5 ml
UDITROL 'A' (Abnormal Serum Control) REF # 070A-010 2x5 ml

REFERENCES

1. Holvey, D.N. ed. : The Merck Mannnal of Diagnosis and Therapy, Merck & Co. Inc., Rahyway, N.J. (1972)
2. Cooper, G.R., CRC Crit Rev. Clin. Lab Sci. 4: 1011 (1973)
3. Tietz, NW, Fundamentals of Clinical Chemistry, WB Saunders Co, Phila. (1970).
4. Trinder, P. J. Clin.Path. 22:246 (1969).
5. Trinder, P. Ann Clin. Biochem. 6:24 (1969)
6. Sunderman, FW. et al, Am J. Clin. Path. 26:1355 (1956)
7. Young, DS. et al, Clin. Chem. 21:1D (1975)

PRODUCT AVAILABILITY

GLUCOSE (OXIDASE) LIQUID
TRINDER/GOD (COLORIMETRIC)

REF # 037L-250	**1 x 250 ml**
REF # 037L-500	**2 x 250 ml**
REF # 037L-1000	**4 x 250 ml**

EC REP
(Authorized representative in the European Community)

mdi Europa GmbH
Wittekamp 30
D-30163 Hannover
Germany

CE