GLUCOSE-L (Single Liquid)

ENZOPAK

(GOD/POD, Enzymatic)

Last update 09-2020

Ref. CC3-GLU.11N, 6x125 ml

INTENDED USE

Reagent kit for quantitative estimation of glucose in serum or plasma.

PRODUCT HIGHLIGHTS

- Low blank.
- End Point formulation with 10 minutes incubation.
- Linearity & Accuracy as per International Standard.
- With lipid clearing factor for accuracy of results.

INTRODUCTION

Conventional methods involved lengthy procedures and were cumbersome.

Glucose-L is based on GOD/POD method (Glucose oxidase / Peroxidase) as described by Trinder and is specific and accurate. The method specifically estimates D-Glucose in serum or plasma.

Further modifications were made to develop a liquid stable Glucose reagent based on the same GOD/POD method.

This has been universally accepted by the customers due to ease in operation and also the reliability of the system.

DIAGNOSTIC SIGNIFICANCE

Blood glucose estimations are generally carried out for the diagnosis and follow up of diabetes mellitus.

In general terms glucose levels less than 50 mg/dl or so are termed as hypoglycemia and more than 200 mg/dl levels are termed as hyperglycemia. Hyperglycemia and Hypoglycemia are also associated with various hormonal disorders e.g. Hormones from Pituitary, Thyroid etc.

PRINCIPLE

Glucose oxidase oxidises the specific substrate, ß -D-glucose, to gluconic acid and generates hydrogen peroxide. Hydrogen-peroxide thus produced is acted upon by peroxidase which transfers oxygen to the chromogen system, 4-aminoantipyrine and phenolic compound. The chromogen system gets oxidised to a red quinoneimine dye. The intensity of colour is directly proportional to the concentration of glucose and is measured photometrically at 505 nm (500-540 nm or with GREEN filter).

$$\text{Glucose} + O_2 + H_2O \xrightarrow{\text{Glucose Oxidase}} \text{Gluconate} + H_2O_2$$

$$H_2O_2 + \text{Phenolic Compound} + \text{4-Aminoantipyrine} \xrightarrow{\text{Peroxidase}} \text{Coloured Complex} + H_2O$$

PRESENTATION

All reagents to be stored at 2-8^{0}C.	No. of Bottles 6 x 125 ml
• Glucose-L (Ready to Use)	6
• Standard (100 mg/dl)	2

FINAL REAGENT COMPOSITION

Active Ingredients	Concentration
• Buffer	100 mmol/L
• GOD	≥ 15000 U/L
• POD	≥ 1000 U/L
• 4 - AAP	0.2 mmol/L

pH 7.4 + 0.5 at 25^0 C

Standard (100 mg/dl)

Also contains non-reactive fillers and Stabilizers.

PREPARATION OF WORKING REAGENT

Glucose-L reagent is ready to use.

PRECAUTION

Glucose is for *IN-VITRO* diagnostic use only.

Reagent Contains Sodium Azide. DO NOT INGEST.

REAGENT STORAGE AND STABILITY

Glucose-L reagent is stable until the expiry date printed on the label of the bottle, when stored at 2-8^{0}C in the original bottle.

SPECIMEN COLLECTION

Blood sample collected with any one of the anticoagulants like fluoride, oxalate, EDTA, heparin or without any of the anticoagulants can be used. To obtain better accuracy and prevent glycolysis, it is necessary to separate serum or plasma as soon as possible.

REACTION PARAMETERS

• Type of Reaction	:	End Point
• Wavelength	:	505 nm (500-540 nm)
• Flowcell Temperature	:	37 ^{0}C
• Incubation	:	10 min. at 37 ^{0}C
• Std. Concentration	:	100 mg/dl
• Sample Volume	:	10 µl (0.01 ml)
• Reagent Volume	:	1.0 ml
• Zero setting with	:	Reagent Blank
• Light Path	:	1.0 cm

TEST PROCEDURE

Pipette Into Test Tubes	BLANK	STANDARD	TEST
Reagent (ml)	1.0	1.0	1.0
Standard (ml)	-	0.01	-
Sample (ml)	-	-	0.01

Mix well and allow to stand for 10 Min. at 37^{0}C. Read absorbance of standard and test at 505 nm (500-540 nm or with GREEN filter) against reagent blank.

The colour of reaction mixture is stable for 2 hours at room temperature, when protected from direct light.

TEST RESULTS

$$\text{Glucose concentration (mg/dl)} = \frac{\text{Absorbance of Test}}{\text{Absorbance of Std.}} \times 100$$

REACTION PARAMETERS

• Type of Reaction	:	Fix Time/Two Point/Initial Rate
• Wavelength	:	505 nm (500-540)
• Flowcell Temperature	:	37^{0}C
• Delay time	:	30 seconds
• Interval time	:	60 seconds
• Measuring time	:	90 seconds
• No. of Readings	:	2
• Standard/Sample Volume	:	10 µl (0.01ml)
• Standard Concentration	:	100mg/dl
• Reagent Volume	:	1.0 ml
• Zero Setting with	:	Distilled Water
• Light Path	:	1.0 cm

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TEST PROCEDURE

Pipette Into Test Tubes	STANDARD	TEST
Reagent (ml)	1.0	1.0
Standard (ml)	0.01	-
Sample (ml)	-	0.01

Mix and aspirate. Record the absorbance of standard (ST) and Test (TS) at 30 seconds (ST_1, TS_1) and again at 90 seconds (ST_2, TS_2) at 505 nm, against distilled water.

TEST RESULTS

$$\text{Glucose concentration (mg/dl)} = \frac{(TS_2-TS_1)}{(ST_2-ST_1)} X\ 100$$

To convert mg/dl to mmol / lit. use the following factor.
1 mmol / lit. = 18 mg/dl
1 mg/dl = 0.056 mmol/lit

LIMITATIONS FOR INTERFERENCE

As per studies carried out for interference. Following results were obtained.

- No Interference from Hemoglobin upto 187.5 mg/dl.
- No Interference from free Bilirubin upto 25.0 mg/dl.
- No Interference from Lipemic (Measured as Triglycerides) upto 1000 mg/dl.

NORMAL VALUES

Fasting : 70-110 mg / dl (3.90-6.11 mmol / lit)
Two Hours Post prandial : upto 140 mg/dl (7.78 mmol/lit)

LINEARITY

This method is linear upto 500 mg/dl. For sample value above 500 mg/dl, dilute the sample suitably with 0.9 % saline & repeat the assay. Apply correction due to dilution to arrive at a final result.

REFERENCES

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