Infinity™
Glucose Oxidase Liquid Stable Reagent

PRODUCT SUMMARY

Stability	:	Until expiry at 2-8°C
Linear Range	:	0-35 mmol/L (0-630 mg/dL)
Specimen Type	:	Serum, plasma or urine
Method	:	Enzymatic Endpoint
Reagent Preparation	:	Supplied ready to use.

IVD

SYMBOLS IN PRODUCT LABELLING

EC REP	Authorized Representative		Temperature Limitation
IVD	For in vitro diagnostic use		Use by/Expiration Date
LOT	Batch code/Lot number		CAUTION. CONSULT INSTRUCTIONS FOR USE.
REF	Catalogue number		Manufactured by
	Consult instructions for use		

INTENDED USE
This reagent is intended for the in vitro diagnostic use in the quantitative determination of glucose in human serum, plasma or urine.

CLINICAL SIGNIFICANCE
The accurate estimation of glucose is important in the diagnosis and management of hyperglycaemia and hypoglycaemia. Hyperglycaemia may occur as a result of diabetes mellitus, in patients receiving glucose containing fluids intravenously, during severe stress and cerebrovascullar accidents. Hypoglycaemia may be the result of an insulinoma, insulin administration, inborn errors of carbohydrate metabolism or fasting.[1] Often in the investigation of these disorders glucose determinations are performed in conjunction with various tolerance tests or stimulation tests. For a more detailed discussion of glucose metabolism the user should refer to a standard text book such as Kaplan.[2]

METHODOLOGY
The glucose oxidase reaction in conjunction with an auxiliary reaction has been widely used for the determination of glucose in biological fluids. Many different auxiliary reactions have been developed in order to improve the overall specificity of the reaction system or retain the inherent specificity of glucose oxidase.[3]
The method utilised in this reagent is based on the hydrogen peroxide indicator reaction which couples 4-aminoantipyrine to a phenolic compound as first proposed by Trinder.[4] This method has been validated in an extensive study by Pennock et al.[5] Pennock compared Trinder's method with six other common methods and found it highly reliable with respect to both accuracy and precision. The method was further shown by Pennock[5] and Sharp[6] and Szasz et al[7] to be resistant to well known interfering compounds such as uric acid, glutathione and creatinine.

1. $\text{Glucose} + O_2 + H_2O \xrightarrow{\textit{Glucose Oxidase}} \text{Gluconic Acid} + H_2O_2$
2. $H_2O_2 + \text{HBA} + \text{4-AAP} \xrightarrow{\textit{Peroxidase}} \text{Quinoneimine dye} + H_2O$

1. Glucose is oxidized by glucose oxidase to gluconic acid and hydrogen peroxide.
2. The hydrogen peroxide reacts in the presence of peroxidase with HBA and 4-aminoantipyrine forming a red quinoneimine dye. The intensity of the color formed is proportional to the glucose concentration and can be measured photometrically between 460 and 560 nm.

Abbreviations
HBA = 4-hydroxybenzoic acid
4-AAP = 4-aminoantipyrine

REAGENT COMPOSITION

Active Ingredients	Concentration
Glucose oxidase	>15 000 U/L
Peroxidase	>100 U/L
4-aminoantipyrine	0.5 mmol/L
4-hydroxybenzoic acid	10 mmol/L
Phosphate buffer	119 mmol/L

Also contains non-reactive fillers and stabilizers.
pH 7.5 ± 0.10 at 20°C

WARNING: Do not ingest. Avoid contact with skin and eyes. If spilt, thoroughly wash affected areas with water. Reagent contains sodium azide which may react with copper or lead plumbing. Flush with plenty of water when disposing. For further information consult the Infinity Glucose Oxidase Reagent Material Safety Data Sheet.

CAUTION: This product contains animal source material. Handle and dispose of this product as if it were potentially infectious.

REAGENT PREPARATION
The reagent is supplied ready to use.

STABILITY AND STORAGE
Prior to use:
When stored refrigerated at 2-8°C the reagent is stable until the expiry date stated on the bottle and kit box label.

Once opened:
Once opened, the reagent is stable until the expiry date stated on the bottle and kit box label when stored refrigerated at 2-8°C.

Indications of Reagent Deterioration:
- Turbidity,
- Reagent Absorbance >0.70 AU at 500 nm, and/or
- Failure to recover control values within the assigned range.

SPECIMEN COLLECTION AND HANDLING
Collection: The stability of glucose specimens is reduced by bacterial contamination and glycolysis. In order to inhibit glycolysis samples should be collected into tubes containing Sodium Fluoride. As soon as possible serum or plasma should be separated from the cells.
Serum: Use non - haemolysed serum.
Plasma: Use EDTA or Heparin.
Urine: If a delay in transport to the laboratory is expected the use of a chemical preservative such as Merthiolate (0.23 mmol/L) is recommended.[8]
Storage: In separated, non-haemolysed serum or plasma, glucose is stable for up to 72 hours at 4°C or as long as 8 hours at 25°C.[2,9.] In the presence of sodium fluoride, glucose is stabilized for as long as 3 days at room temperature.[10] For long term storage samples should be placed in sealed containers and frozen at -10°C.[11] Urine samples are stable for 7 days at 4°C.[4]

ADDITIONAL EQUIPMENT REQUIRED BUT NOT PROVIDED
- A clinical chemistry analyser capable of maintaining constant temperature (37°C) and measuring absorbance at 500 nm, 460 nm or 560 nm.
- If required, pipettes for accurately dispensing measured volumes.
- Analyser specific consumables, eg: sample cups.
- Normal and abnormal assayed control material.
- Calibrator or a suitable aqueous Glucose standard.

ASSAY PROCEDURE
The following system parameters are recommended. Individual instrument applications are available upon request from the Technical Support Group.

SYSTEM PARAMETERS

Temperature	37°C
Primary Wavelength	500 nm (460 nm, 560 nm)
Secondary Wavelength	600 - 660 nm
Assay Type	Endpoint
Direction	Increase
Sample : Reagent Ratio	1 : 150
e.g.: Sample Vol	3 µL
Reagent Vol	450 µL
Incubation Time	5 minutes
Reagent Blank Limits	Low 0.00 AU
(500 nm, 1cm lightpath)	High 0.70 AU
Linearity	0-35 mmol/L (0-630 mg/dL)
Analytical Sensitivity	0.035 ΔAbs per mmol/L
(500 nm, 1cm lightpath)	0.002 ΔAbs per mg/dL

CALCULATIONS
Results are calculated, usually automatically by the instrument, as follows:

$$\text{Glucose} = \frac{\text{Absorbance of Unknown}}{\text{Absorbance of Calibrator}} \times \text{Calibrator Value}$$

Example:
Absorbance of calibrator = 0.46
Absorbance of unknown = 0.10
Value of calibrator = 13.2 mmol/L (238 mg/dL)

$$\text{Glucose} = \frac{0.10}{0.46} \times 13.2 = 2.76 \text{ mmol/L}$$

$$\text{Glucose} = \frac{0.10}{0.46} \times 238 = 51 \text{ mg/dL}$$

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For urine specimens the results must be multiplied by the dilution factor and 24 hour collections by the volume in liters.

Urine Glucose (mmol/24 hours)	=	Glucose Result (mmol/L)	x	Dilution Factor	x	Volume (L)

Example:

Glucose result	=	0.7 mmol/L (12.6 mg/dL)
Dilution of Urine	=	Neat
24 Hour volume of urine	=	0.95 Liters

Urine Glucose	=	0.7 x 1 x 0.95	=	0.67 mmol/24 hours
Urine Glucose	=	12.6 x 1 x 0.95	=	11.97 mg/24 hours

NOTES

1. The reagent and sample volumes may be altered proportionally to accommodate different spectrophotometer requirements.
2. Specimens with glucose values above 35 mmol/L (630 mg/dL) should be diluted with isotonic saline and reassayed. Multiply results by the dilution factor.
3. Unit conversion: mmol/L x 18 = mg/dL.
4. Avoid direct sunlight.

CALIBRATION

Calibration is required. An aqueous standard or serum based calibrator, with an assigned value traceable to a primary standard (eg NIST or IRMM) is recommended. For calibration frequency on automated instruments, refer to the instrument manufacturers specifications.

However, calibration stability is contingent upon optimum instrument performance and the use of reagents which have been stored as recommended in the stability and storage section of this package insert. Recalibration is recommended at anytime if one of the following events occurs:-

- The lot number of reagent changes.
- Preventative maintenance is performed or a critical component is replaced.
- Control values have shifted or are out of range and a new vial of control does not rectify the problem.

QUALITY CONTROL

To ensure adequate quality control, normal and abnormal control with assayed values for this methodology should be run as unknown samples:-

- At least once per day or as established by the laboratory.
- When a new bottle of reagent is used.
- After preventative maintenance is performed or a critical component is replaced.
- With every calibration.

Control results falling outside the upper or lower limits of the established ranges indicate the assay may be out of control.

The following corrective actions are recommended in such situations:-

- Repeat the same controls.
- If repeated control results are outside the limits, prepare fresh control serum and repeat the test.
- If results are still out of control, recalibrate with fresh calibrator, then repeat the test.
- If results are still out of control, perform a calibration with fresh reagent, then repeat the test.
- If results are still out of control, contact Technical Services or your local distributor.

LIMITATIONS

1. Studies to determine the level of interference from haemoglobin, bilirubin and lipaemia were carried out. The following results were obtained:
   Haemoglobin: No interference from haemoglobin up to 750 mg/dL.
   Bilirubin: No interference from bilirubin up to 770 µmol/L (45 mg/dL).
   Lipaemia: No interference from lipaemia, measured as triglycerides up to 23 mmol/L (2000 mg/dL).
2. For a more comprehensive review of factors affecting glucose assays refer to the publication by Young.[12]

EXPECTED VALUES

Fasting serum: [13]	4.11 - 5.56 mmol/L (74 - 100 mg/dL)
Urine:[13]	0.06 - 0.83 mmol/L (1 - 15 mg/dL)

For the diagnosis of diabetes, Impaired Fasting Glucose (IFG) or Impaired Glucose Tolerance (IGT) the W.H.O. recommend the following criteria:[14]

Diabetes

Fasting plasma glucose	≥7.0 mmol/L (≥126 mg/dL)
2 hrs after glucose load	≥11.1 mmol/L (≥200 mg/dL)

IFG

Fasting plasma glucose	6.1-6.9 mmol/L (110-125 mg/dL)

IGT

Fasting plasma glucose	≤7.0 mmol/L (≤126 mg/dL)
2 hrs after glucose load	7.8-11.0 mmol/L (140-199 mg/dL)

PERFORMANCE DATA

The following data was obtained using the Infinity Glucose Oxidase Liquid Stable Reagent on a well maintained automated clinical chemistry analyser. Users should establish product performance on their specific analyser used.

IMPRECISION

Imprecision was evaluated over a period of 20 days using two levels of commercial control and following the NCCLS EP5-T procedure.[15]

Within Run:	LEVEL I	LEVEL II
Number of data points	80	80
Mean (mmol/L / mg/dL)	4.99 / 90	17.31 / 312
SD (mmol/L / mg/dL)	0.12 / 2.2	0.18 / 3.2
CV (%)	2.4	1.0
Total:	LEVEL I	LEVEL II
Number of data points	80	80
Mean (mmol/L / mg/dL)	4.99 / 90	17.31 / 312
SD (mmol/L / mg/dL)	0.26 / 4.7	1.01 / 18.2
CV (%)	5.2	5.8

METHOD COMPARISON

Comparison studies were done using another commercially available glucose oxidase reagent as a reference. Normal and abnormal patient samples were assayed in parallel. The results were compared be least squares regression and the following statistics were obtained.

Number of sample pairs	60
Range of sample results	3.1 - 17.0 mmol/L (56-306 mg/dL)
Mean of reference method results	6.94 mmol/L (125 mg/dL)
Mean of Glucose Infinity results	6.69 mmol/L (120 mg/dL)
Slope	1.002
Intercept	0.157 mmol/L (2.8 mg/dL)
Correlation coefficient	0.998

LINEARITY

When run as recommended the assay is linear between 0 and 35 mmol/L (0 - 630 mg/dL). Linearity on automated instruments may vary from the quoted value. It is recommended that the user refer to the appropriate Thermo instrument application for the instrument specific linearity claim.

ANALYTICAL SENSITIVITY

When run as recommended the sensitivity of this assay is 0.035 ΔAbs per mmol/L or 0.002 ΔAbs per mg/dL (1cm light path, 500 nm).

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Fisher Diagnostics
a division of Fisher Scientific Company, LLC
a part of Thermo Fisher Scientific Inc.
Middletown, VA 22645-1905 USA
Phone: 800-528-0494
540-869-3200
Fax: 540-869-8132

EC REP WMDE
Bergerweg 18
6085 AT Horn
The Netherlands

CE

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REF

Reorder Information

Catalogue No.	Configuration
TR15221	2 x 125 mL