



GLUCOSE (GOD-POD Method, End Point)

Intended Use

The reagents are used for the quantitative determination of Glucose in serum or plasma. For in-vitro diagnostic use only.

Introduction

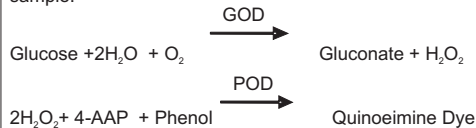
Glucose is the reducing monosaccharide that serves as the principal source of cellular energy in the body. It enters into the cell under the influence of insulin and undergoes a series of chemical reactions to produce energy. Lack of insulin or resistance to its action at the cellular level causes diabetes. Therefore, in diabetes mellitus the blood glucose level are very high. However, high blood glucose level is also observed in the pancreatitis, pituitary or thyroid dysfunction, renal failure and liver disease whereas low glucose level is associated with starvation, hyperinsulinaemia, neoplasms or insulin induced hypoglycemia.

Method

GOD-POD method, End Point.

Principle

Glucose is oxidized by glucose oxidase(GOD) to produce gluconate and hydrogen peroxide. The hydrogen peroxide is then oxidatively coupled with 4 amino- antipyrine(4-AAP) and phenol in the presence of peroxidase(POD) to yield a red quinoeimine dye that is measured at 505nm. The absorbance at 505 nm is proportional to concentration of glucose in the sample.



Absorbance of the colored solution is directly proportional to the glucose concentration, when measured at 505nm.

Reagent Composition

Reagent 1:

Glucose Oxidase	20000 u/l
Peroxidase	1200 u/l
4-AAP	0.246 mmol/l

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Reagent 2:

Glucose Standard	100 mg/dl
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Precautions

Following precautions should be taken:

- Avoid ingestion, do not pipette by mouth.
- Avoid contact with skin and eyes. If spilled, thoroughly wash affected area with water.
- Flush with plenty of water while disposing.

Reagent Storage and Stability

Unopened Reagent 1 is stable till expiry mentioned on the label when stored at 2-8°C.

Standard Reagent 2 is stable till expiry as mentioned on the label when stored at 2-8°C.

Note: On request, Reagent 3 (Glucose: 300 mg/dl) & Reagent 4 (Glucose: 500 mg/dl) can be provided for linearity check with Reagent 2 (Glucose: 100 mg/dl-Standard).

Reagent Preparation

Reagents are ready for use.

Reagent Deterioration

Reagent should be clear. Turbidity and/or precipitation may be because of reagent deterioration.

Sample Collection and Storage

Unhaemolysed serum or plasma can be used for the testing. Anti-coagulants like EDTA and heparin can be used. Preferable sodium fluoride should be used as anti-coagulant. It is recommended to use freshly collected samples for assay. Separated plasma samples can be stored for 3 days at 2-8°C.

General Assay Parameters

Mode	End Point
Wavelength (nm)	505
Wavelength Range Usable(nm)	500-550
Blank with	Reagent
Sample Volume (µl)	5/10
Reagent R1 (µl)	500/1000
Incubation Time (min)	15 /7
Incubation Temperature (°C)	RT/37
Normal Low (mg/dl)	70
Normal High (mg/dl)	110
Linearity (mg/dl)	Upto 500
Standard Conc. (mg/dl)	100
Units	mg/dl

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Procedure

One reagent blank and one standard are sufficient for each assay series.

Pipette into test tubes:

Particulars	Blank	Standard	Sample
Reagent 1	1000µl	1000µl	1000µ
Dist. Water	10µl	-	-
Reagent 2	-	10µl	-
Sample	-	-	10µl

Mix well & incubate for 15 min at room temperature or 7 min at 37°C. Measure the absorbance of standard (Astd) and sample (Asample) against reagent blank at 505 nm

Calculation

Glucose concentration in the sample can be calculated using the following formula:

$$\text{Glucose} = \frac{\text{Absorbance of Sample}}{\text{Absorbance of Standard}} \times \text{Conc. of Std. (mg/dl)}$$

Example: If the absorbance of sample is 0.200 and the absorbance of standard is 0.18. The calculation shall be

$$\frac{0.200}{0.180} \times 100 = 111.1 \text{ mg/dl}$$

If the glucose concentration exceeds 500 mg/dl, dilute the sample with normal saline and repeat the assay. The reportable results in this case shall be calculated by multiplying the results obtained with dilution factor.

Reference value

70 –110 mg/dL

Limitations

1.The reagent and sample volumes can be altered proportionately so that the sample:reagent ratio remains same.

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2. Hemolytic and lipemic samples may result in false elevated results. To avoid false results sample blank may be used as mentioned below:

- Add 10µl of serum sample to 1000µl of DI water and read absorbance at 505nm.
- Subtract the absorbance obtained as above, from the absorbance of test. Use this corrected absorbance for calculation.
- Reagents are sensitive to light and temperature.

Quality Control

The patient results obtained for each batch can be validated by using normal and abnormal control sera with assayed values for glucose.

Performance

Linearity Limit: 500mg/dl

Precision:

Within run

Control	Control 1	Control 2
No. of samples	20	20
Mean (mg/dl)	95.7	305.1
S.D.	1.14	1.35
C.V. %	1.19	0.44

Between run

Control	Control 1	Control 2
No. of samples	60	60
Mean (mg/dl)	95.8	304.8
S.D.	1.25	1.85
C.V. %	1.30	0.61

References

1. Juaristi, Eusebio; Gabriel Cuevas (1995). The Anomeric Effect. CRC Press. pp. 9– 10. ISBN 0849389410 <http://wapedia.mobi/en/Glucose?t=9>.

2. Kirschner, K.N. Woods, R.J. (2001). "Solvent interactions determine carbohydrate conformation". Proc. Natl. Acad. Sci. USA. 98 (19):10541–10545. doi:10.1073/pnas.191362798.PMID11526221 <http://en.wikipedia.org/wiki/Glucose>
3. National Committee for Clinical Laboratory Standards. Approved Guideline, NCCLS publication C28-A, Villanova, PA (1994). www.tecodiag.com/Admin/pdf/356_G513_500B.pdf
4. McMurry, John (1988). Organic Chemistry. Brooks/Cole. pp. 866. ISBN 0534079687. en.wikipedia.org/wiki/Glucose

Pack Presentation

Product Code /Catalogue No.	Pack Size*	Reagent 1	Reagent 2
KGGLU104.1.1	5x100ml	5x100ml	1x3ml

* Pack size may vary on market demand.

Revision No: (Ver: KGGLU104.1/1)

Date of Issue:1st April 2010

Symbols

Following symbols are used in the labeling of KEE GAD kits:



Catalogue No.



Batch No.



CE Mark



Read instructions



In Vitro Diagnostics



Storage temperature



Expiry Date



Content



Product Name



Manufactured By



Manufactured by:

KEE GAD Biogen Pvt. Ltd.

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