# Manual Procedure

Automated procedure on request

MEDICHEM MIDDLE EAST
Clinical Chemistry Reagents
Liquid Stable Reagents

**Cat. No. 12380** R 2 x 50 ml
**For 100 tests**

**Cat. No. 12381** R 6 x 50 ml
**For 300 tests**

**Cat. No. 12383** R 4 x 250 ml
**For 1000 tests**

# Glucose GOD/PAP

Enzymatic colorimetric test

***Liquid Reagent***

## Test Principle

Enzymatic colorimetric determination of glucose according to the following reactions:

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

$$2\,H_2O_2 + \text{Phenol} + \text{4-Aminoantipyrine} \xrightarrow{Peroxidase} \text{4-(p-bezoquinone-mono-imino)phenazone} + 4H_2O$$

Glucose is oxidized by glucose oxidase to gluconate and hydrogen peroxide. In the presence of peroxidase, phenol reacts with 4-AAP and hydrogen peroxide to produce a Quinonimin dye, the intensity of the colour produced measured at 505 nm, is proportional to the concentration of glucose in the sample.

## Concentrations in the test

| **Reagent** | | |
|---|---|---|
| Phosphate buffer pH = 7.40 | 120.0 | mmol/L |
| Phenol | 10.0 | mmol/L |
| Glucose oxidase | ≥ 30 | KU/L |
| Peroxidase | ≥ 5 | KU/L |
| 4-Aminoantipyrine | 0.3 | mmol/L |
| Preservative, stabilizer. | | |

**Standard :** The Concentration as indicated on vial.

## Stability of reagent

**Reagent :** *liquid, ready to use.*
The reagent is stable up to expiry date given on the label when stored at +2 ➔ +8°C.

**Note:** The reagent should not be used if it has developed turbidity or other evidence of microbial growth.

## Specimen collection and handling

1. Fresh, non-hemolyzed serum.
2. Non-hemolyzed plasma collected on oxalate, citrate, EDTA, fluoride or heparin.
3. Cerebrospinal fluid (CSF).
4. Glucose in serum is stable for 24 hours at 20 - 25°C and 7 days at 2 - 8°C, after addition of a glycolysis inhibitor (NaF, KF).
5. Serum and plasma must be separated from the red cells promptly to prevent glycolysis. Glucose will decrease approximately 7 % per hour when left in contact with red cells.
6. Use fluoride plasma in case of a delay in analysis.

## Calibrator / Standard

MediCal U Cat. No. 15011
Glucose STD. Cat. No. 16111

## Quality control

Meditrol N Cat. No. 15171
Meditrol P Cat. No. 15181

## Procedure

| | |
|---|---|
| Wavelength | Hg 546 nm (492 - 550 nm) |
| Spectrophotometer | 505 nm |
| Cuvette | 1 cm light path |
| Temperature | 37°C / 20 - 25 °C |
| Measurement | against blank |
| Reaction | end point |

## Assay

| | Blank | Calibrator / Standard | Sample |
|---|---|---|---|
| Distilled water | 10 µl | -- | -- |
| Calibrator/ Standard | -- | 10 µl | -- |
| Sample | -- | -- | 10 µl |
| Reagent | 1000 µl | 1000 µl | 1000 µl |
| Mix, incubate for 8 min. at 37 °C or 12 min. at 20 - 25 °C. Read the absorbance (A). The final color is stable for 1 hour. | | | |

## Calculation

$$\text{Conc.}_{\text{Glucose}}\ (\text{mg/dl}) = \frac{A_{\text{Sample}}}{A_{\text{Cal./STD.}}} \times \text{Conc.}_{\text{Cal./STD.}}\ (\text{mg/dl})$$

$$\text{mmol/L} \underset{0.0555\ \times}{\overset{\times\ 18}{\rightleftharpoons}} \text{mg/dl}$$

## Linearity

Up to 500 mg/dl (27.75 mmol/L).
If the result exceeds 500 mg/dl, repeat the test using diluted sample (1+2) with sodium chloride solution (0.9 %) and multiply the result by 3.

## Interferences

1. Bilirubin up to 20 mg/dl and hemoglobin up to 500 mg/dl have both been found to exhibit negligible interference (< 3 %) in this assay.
2. Grossly lipemic samples may cause falsely elevated glucose values.
3. A number of drugs and substances affect glucose results. see Young, et al.[2]

## Precaution

The reagent contains sodium azide. Don't swallow. Avoid any contact with skin and mucous membranes. Sodium azide may react with lead and copper plumbing to form explosive metal azides. Upon disposal, flush with large amounts of water to prevent azide build up.

## Reference range

| | | | |
|---|---|---|---|
| Serum | Adults | 75 - 115 | mg/dl |
| Plasma (venous) | Adults | 55 - 115 | mg/dl |
| CSF | Adults | 50 - 70 | mg/dl |

## References


1. Trinder P., Ann. Clin., Biochem, 6, (1969), 24.
2. Young, DS., Effects of Drugs on Clinical Laboratory Tests, fifth edition 2000, AACC Press, Washington, D.C.
3. Thoma Ed. Med. Veriagsgesellschaft, Marburg.
4. Young et. al., Clin. Chem. 21 :1D, 1975.