

Experiment

Determination of Blood Sugar

Aim

To determine the concentration of glucose (blood sugar) in a given blood sample using the Glucose Oxidase–Peroxidase (GOD-POD) method.

References

K.K Pillai and J.S Qadry Biochemistry and clinical pathology. CBS Publication and Distributors Pvt. LTD, New Delhi, First edition, 127-128.

Requirements

- **Glassware Requirement:-** Test tubes, Micropipette, Test tube stand, Spectrophotometer/Colorimeter, Water bath, Centrifuge.
- **Chemical Requirement:-** Glucose oxidase reagent, Phenol reagent, 4-Aminophenazone, Standard glucose solution, Distilled water, Blood serum sample.

Principle

Glucose oxidase oxidizes glucose to gluconic acid and hydrogen peroxide. In the presence of peroxidase, hydrogen peroxide reacts with phenol and 4-aminophenazone to form a pink coloured quinoneimine complex. The colour intensity measured at 505 nm is directly proportional to the glucose concentration.

Procedure

Label three test tubes as Blank (B), Standard (S), and Test (T).

1. Add 1.0 ml working reagent to all tubes.
2. Add 10 μ l distilled water to Blank.
3. Add 10 μ l standard glucose to Standard tube.
4. Add 10 μ l serum sample to Test tube.
5. Mix well and incubate at 37°C for 10 minutes.
6. Measure absorbance of Standard and Test against Blank at 505 nm.

Experimental Data (Example)

Tube	Working Reagent (ml)	Sample/Standard (μ l)	Absorbance
Blank	1.0	Distilled water 10 μ l	0.00
Standard	1.0	Standard 10 μ l	0.50
Test	1.0	Serum 10 μ l	0.65

Calculation (Example)

Blood Glucose (mg/dL) = (Absorbance of Test / Absorbance of Standard) × 100

$$\begin{aligned} &= (0.65 / 0.50) \times 100 \\ &= 130 \text{ mg/dL} \end{aligned}$$

Result

The blood glucose concentration in the given sample is 130 mg/dL.

Normal Values

- **Fasting:** 70–110 mg/dL
- **Post-prandial:** 110–140 mg/dL