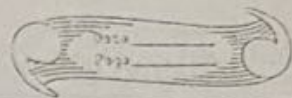


Glucose



* Sample collection & storage:



→ Individual \bar{c} (1) hematocrit, fasting whole blood glucose conc. 10 to 12 ~~mg~~%. lower than plasma.

→ Both serum & plasma can be used for glucose estimation.

→ POCT device uses to measure glucose conc in the plasma phase.

→ Glycolysis ↓es s. glucose by 5 to 10 mg/dl in 1 hr. in (2) uncentrifuged coagulated blood at room temperature.

→ Rate is higher if (1) leucocytosis
(2) Bact. contamination.

leucocytosis may lead to ↓ or \approx 65 mg/dl of glucose after 1 to 2 hrs in -nce of glycolytic inhibitors

→ Separated non hemolyzed sterile serum stable glucose conc. as long as 8 hrs at 25°C.

→ Even the plasma free from cell



may contain leukocyte after mod. centrifugation



that also metabolize glucose

* Glycolytic Inhibitors :-

→ NaF :

↳ Inhibits glycolysis

↓
glucose is stabilized for 3 days
at room temp.

↓
It inhibits activity of endase

↓
By forming complex \bar{c} Mg^{2+} & P_i

↓
c interferes interaction enzyme
& substrate.

→ Weak anticoagulant → ~~it~~ binds \bar{c} Ca^{2+}

↓
So used in combinⁿ \bar{c} oxalate.

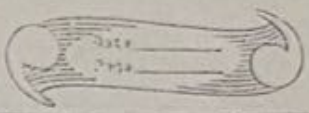
→ Fluorides inhibits urease activity

↓
So not useful for determination of
urea

→ ~~Fluoride~~

→ Glycolytic activity of Fluoride has
late onset ↓

So glycolysis may occur in 1st hr
after collection of sample even in
Fluoride vacuttes.



(b) ^{sod.} Iodoacetate

↓
also inhibit glycolysis. → when want to measure Urea & glucose → Iodoacetate can be use as preservative.

→ In 24 hrs collection of urine

↓
glucose may be preserve. By adding 5 ml of glacial acetic acid to the container

↓
So pH B/w 4 to 5

↓
Inhibits Bact. activity.

Other preservative for urine ?

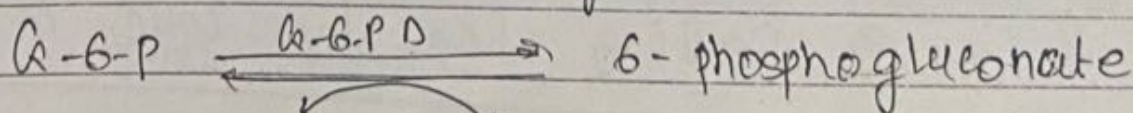
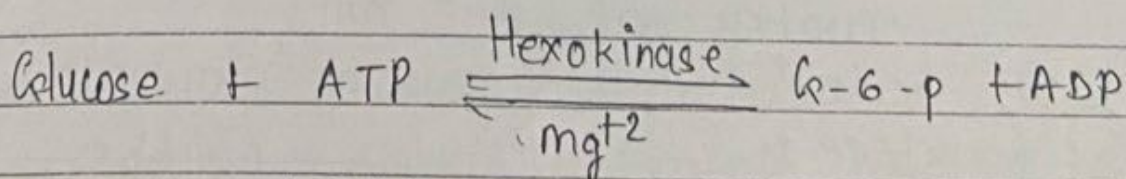
↓
Sod. Benzoate

Sod. Nitrate.

→ Urine should be stored at 4°C during collection.

* Methods :-

(1) Hexokinase methods :-



NADP
(constant) (NADPH + H⁺) → measured at 340nm

* Ref.

Interference :-

① Hb: $> 0.5 \text{ mg/dl}$: interferes.

↓
② B^{12} phosphate esters & enzymes released from RBC
↓
interfere in the assay.

② Drugs

③ Bilirubin

④ Lipemia:

↳ $\text{TG} > 50 \text{ mg/dl}$

↓
+ve interference.

↓
Sample Blanking

↓
to eliminate lipemic & icteric interference.

↓
prepared by 10 μl sample + isotonic saline

↓
absorbance taken against water at 340 nm.

⑤ Fructose:

→ (ii) Fasting has low conc. of fructose

Date _____
Page _____

↓

Ingestion of sucrose 4 gm/kg

↓

↑ S. Fructose \bar{c} to 10 mg/dl \bar{c} in 1 hrs, remain high for 2 hrs.

↓

So during QTT, should not administer any fructose.

* Mode of reaction :-

→ End point / equilibrium assay

* Reagent may also contain substances that reacts \bar{c} coenzyme +nt in the reagent

↓

they can be detected by taking reagent blank

↓

- IF reagent blank at 340nm is > 0.35

↓

they are unsuitable for use.

② Reference method :

→ Serum / plasma deproteinated by adding solution Barium hydroxide & zinc sulfate

↓

Supernatant mixed \bar{c} reagent
↓

method is Based on Hexokinase,
 α -G.P.D & NADH (NOT NADPH)

→ Calibrators & Blank carried through
entire process.

② Glucose Oxidase Methods :-