TOTAL PROTEIN KIT

(Biuret method)

For the determination of Total Proteins in serum and plasma.

(For Invitro Diagnostic Use Only)

Summary

Proteins are constituents of muscle, enzymes, hormones and several other key functional and structural entities in the body. They are involved in the maintenance of the normal distribution of water between blood and the tissues. Consisiting mainly of albumin and globulin the fractions vary independently and widely in diseases.Increased levels are found mainly in dehydration. Decreased levels are found mainly in malnutrition, impared synthesis, protein losses as in hemorrhage or excessive protein catabolism.

Principle

Proteins, in an alkaline medium, bind with the cupric ions present in the biuret reagent to form a blue-violet coloured complex. The intensity of the colour formed is directly proportional to the amount of Proteins present in the sample.

Proteins + Cu^{++} ⟶ Blue Violet Coloured Complex

Normal reference values

Serum & Plasma : 6.0 - 8.0 g/dl

It is recommended that each laboratory establish its own normal range representing its patient population.

Contents	**150 ml**	**2 x 150ml**	**3 x 150 ml**
Carton 1			
L1 : Biuret Reagent	150 ml	2 x 150 ml	3 x 150 ml
Carton 2			
S : Protein Standard (8 g/dl)	5 ml	5 ml	5 ml

Storage / stability

Carton 1 : Biuret Reagent is stable at R.T. till the expiry mentioned on the label.
Carton 2 : Protein Standard is stable at 2-8^{0}C till the expiry mentioned on the label.

Reagent Preparation

Reagents are ready to use. Protect from bright light.

Sample material

Serum or plasma. Proteins are reported to be stable in the sample for 6 days at 2 - 8^{0}C.

Procedure

Wavelength / filter : 550 nm (Hg 546 nm) / Yellow - Green
Temperature : R.T. / 37^{0}C
Light path : 1 cm

Pipette into clean dry test tubes labeled as Blanks (B), Standard (S), and Test (T) :

Addition Sequence	**B** (ml)	**S** (ml)	**T** (ml)
Biuret reagent (L1)	1.0	1.0	1.0
Distilled water	0.02	-	-
Protein Standard (S)	-	0.02	-
Sample	-	-	0.02

Mix well and incubate at 37^{0}C for 10 min.or at R.T. for 30 min. Measure the absorbance of the Standard (Abs.S), and Test Sample (Abs.T) against the Blank, within 60 Min.

Calculations

$$\text{Total Proteins in g/dl} = \frac{\text{Abs.T}}{\text{Abs.S}} \times 8$$

Linearity

This procedure is linear upto 15 g/dl. If values exceed this limit, dilute the sample with distilled water and repeat the assay. Calculate the value using the proper dilution factor.

Note

Do not use if the reagent shows turbidity or black precipitates.

References

Gornall, A.G., etal, (1949) Biol. Chem. 177 : 751
Doumas, B.T., (1975) Clin Chem. 21 : 1159

System Parameters

Reaction	: End Point	**Interval**	: ---
Wavelength	: 550 nm	**Sample Vol.**	: 0.02 ml
Zero Setting	: Reagent Blank	**Reagent Vol.**	: 1.00 ml
Incub. Temp.	: 37°C / R.T.	**Standard**	: 8 g/dl
Incub. Time	: 10 min./ 30 min.	**Factor**	: ---
Delay Time	: ---	**React. Slope**	: Increasing
Read Time	: ---	**Linearity**	: 15 g/dl
No. of read.	: ---	**Units**	: g/dl

Coral Clinical Systems

Gitanjali, Dr. Antonio Do Rego Bagh,
Bambolim Complex P.O. GOA - 403 202, INDIA